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STUDIES ON HETEROCARYOSIS IN ASPERGILLUS AND PENICILLIUM*

SEIZO TSUDA

(Accepted for Publication, July 10, 1955)

In many species of Aspergillus and Penicillium the development of perithecia occurs very rarely in nature. They usually propagate by conidia. But a phenomenon analogous to hybridization has been observed in them, in which one or more nuclei migrate from one mycelium into another and produce the heterocaryon, i.e. hyphal cells which contain two or more genetically different kinds of nuclei, whose numerical balance is fixed so long as the environment of the mycelium remains constant. This system seems to be universal in the Fungi imperfecti and also widespread, side by side with sexual reproduction, in most heterothallic fungi. The heterocaryotic condition may, of course, arise as a consequence of mutation in one or more of the several nuclei of a hyphal cell.

It has been demonstrated by Pontecorvo (1944, 1949, 1953) that different mutants of *Penicillium notatum*, *Penicillium chrysogenum* and *Aspergillus niger* will form heterocaryons with a wild-type phenotype. Heterocaryosis in *Aspergillus*, *Penicillium* and *Neurospora* has been studied among others by Hansen (1938), Gossop *et al.* (1940), Beadle and Coonradt (1944), Lindegren and Andrews (1945), Sakaguchi and Ishitani (1952) and Tsuda (1952, 1953).

The present experiments were carried out with a heterocaryotic strain or Aspergilla maidus; futher more, heterocaryons were artificially induced on synthetic agar media between two varieties of Aspergillus awamori and between two ultra-violet-ray mutants of Penicillium chrysogenum.

MATERIALS AND METHODS

The following cultures were used in this study: a heterocaryotic strain of Aspergillus condidus, two varieties of Aspergillus awamori and two ultra-violet-ray induced mutants from a penicillin producing strain, namely, Penicillium chrysogenum Q 176. green spored and producing yellow pigment, which had been cultured in our laboratory. Strain No. 5-9 of Aspergillus awamori had light brown spores and the other strain, No. N-19, dark brown spores. Two Penicillium chrysogenum mutants were used, one, UY-3S, which formed jagged colonies and was producing large amounts of yellow substances, sorbicillin and penicillinic acid, like the wild type, while the other, UW-1S, formed round and colourless colonies, but the colour of spores of both mutants was equally white.

^{*}Contributions from the National Institute of Genetics, Japan, No. 111

Stock cultures of these fungi were grown on agar medium, and the experimental cultures on agar medium after Czapek-Dox, modified by the addition of 1 g of asparagin per 1 liter. The composition of the experimental medium was as follows: sucrose, 30 g; NaNO₄, 2 g; KH₂ PO₄, 1 g; KCI, 0.5 g; MgSO₄. 7H₂O, 0.5 g; FeSO₄, 0.01 g; asparagin, 1 g; agar, 20 g per liter distilled water.

EXPERIMENTAL RESULTS AND DISCUSSION

The author found in the fall of 1951 a case of heterocaryosis in Aspergillus candidus and obtained a heterocaryotic strain. From this heterocaryon a morphologically different strain was segregated. A colony arising from the heterocaryon on a solid medium was divided into two patches, one heterocaryotic, and the other a homocaryotic patch, as seen from figure 1. The new strain was cultured repeatedly as many as ten times, and found to retain its morphological characteristics, no further segregation occurring. Accordingly, it was assumed that this strain had become homocaryotic.

In the newly segregated homocaryotic patch, the conidiophores were found to be relatively shorter and the conidia somewhat smaller than in the original heterocaryon. It has been noticed that the heterocaryotic strain formed a lamella, and the homocaryotic strain formed a hexenring. The spores were stained according to Robinow's Giemsa method after fixation with osmium tetroxide vapor and hydrolyzation by n.HCl at 60°C. for 7 minutes. Observation of the preparations has revealed that the spores of the heterocaryotic strain contained many nuclei, not less than five, while their number in the spores of the homocaryotic strain usually was only two or three.

In the case of Aspergillus awamori, two colonies derived from two different strains, one No. 5-9 and the other No. N-19 both strains belonging to two varieties of Aspergillus awamori, were grown on solid agar medium starting from inoculi far apart from each other. Hyphal fusion or anastomosis was observed when the colonies met. It occurred between branches of the hyphae which were derived from the two original parental strains as seen from figure 2.

That nuclei from one hypha can migrate into another, following hyphal fusion, has been shown again and again in many species of Fungi imperfecti, as well as in species having a sexual stage. The details of such nuclear migrations, however, remain totally obscure.

The heterocaryon had on surface cultures a growth rate almost equal to that of the parental two strains as seen from figure 3, and the colour of the conidia was about intermediate.

Furthermore, the author has succeeded in inducing a heterocaryon on a synthetic agar medium in two ultra-violet-ray mutants of Penicillium chrysogenum. The wild type of Penicillium chrysogenum Q 176 formed jagged colonies and produced a large amount of yellow coloured substances, namely sorbicillin and penicillinic acid and was green spored. Two mutants induced from the wild type by ultra-violet irradiation were examined. One of these, UY-3S, formed

jagged colonies and produced a large amount of yellow coloured substance, and the other mutant, UW-1S, formed round and colourless colonies. The conidia were whitish in both strains and hardly distinguishable. Two colonies of these two *Penicillium* mutants were grown on solid agar medium starting from inoculi far apart from each other in the same way as in *Aspergillus awamori*, and the development of a heterocaryon was observed when these two colonies established a contact with each other. The conidia produced by the heterocaryon were green and contained yellow substances just as the wild type.

In Aspergillus and Penicillium which mostly produce conidia with one nucleus, the formation of conidia from a heterocaryon automatically leads to segregation of nuclei of different kinds. The number of nuclei in a hyphal cell usually reaches more than a dozen per cell. Accordingly the heterocaryon between the two whitespored mutants will carry one of the two kinds of "white" nuclei to each conidium. All conidia produced by this heterocaryon were green like those of the wild type. It is assumed that the pigment was determined by the heterocaryotic conidiophore and not by the kind of nucleus segregated in each conidium.

A similar situation has been described by Pontecorvo as non-autonomous gene action. His assumption is that "if the two different coloured spore mutants were recessive and nonallelomorphic, their dominant alleles being necessary for the production of two diffusible substances, we should expect the colour of every conidium to be green irrespective of which kind of nucleus was segregated into it."

Figure 5 shows that the character of the colonies derived from this heterocaryon showed a continuous variation between the two parental mutants, UY-3S, (a) which forms jagged colonies and produces a large amount of yellow substances, and UW-1S (h), which forms round and colourless colonies. The presence of colour substances and jaggedness of colony, however, were closely associated.

Accordingly, the production of yellow substances, sorbicillin and penicillinic acid, was decreasing in the direction from (a) to (h) in the figure.

As the number of nuclei in hyphal cells of *Penicillum* usually reaches more than a dozen per cell, it may be reasonably assumed that the continuous variation in the characters of the heterocaryon as stated above resulted from the varying proportion of the nuclei contributed by the two mutants.

SUMMARY

The author investigated a case of heterocaryosis in Aspergillus candidus, and artificially induced heterocaryon on synthetic agar media between two varieties of Aspergillus awamori and two induced ultra-violet-ray mutants of Penicillium chrysogenum.

From the heterocaryon of Aspergillus candidus a morphologically different strain was segregated. The colony arising from the hetero-

caryon on a solid medium segregated into two patches. The segregant was cultured repeatedly, and found to retain its morphological characteristics, no further segregation occurring. Accordingly, a homocaryotic strain was established. In the newly segregated homocaryon, the conidiophore was found to be relatively short, and the size of the conidia somewhat smaller than in the heterocaryon. The number of nuclei in these spores was examined after Robinow's Giemsa staining method.

The heterocaryon between two varieties of Aspergillus awamori, 5-9 and N-19, was intermediate between the two parental varieties so far as colour and form of the colony are concerned.

A heterocaryon in *Penicillium chrysogenum* was induced between two ultra-violet-ray mutants, UY-3S, with jagged colonies producing a large amount of yellow substances, sorbicillin and penicillinic acid and UW-1S which formed round and colourless colonies, but the colour of the spores of both strains was equally white and indistinguishable. The conidia produced by the heterocaryon were green and contained yellow substances just as the wild type, *Penicillium chrysogenum* Q 176 dose.

Variation of the colony characters in the heterocaryotic strains was continuous covering the whole range between the two parents, the presence of colour substances and the jaggedness of colony, however, being closely associated.

As the number of nuclei in hyphal cells of *Penicillium* usually reaches more than a dozen per cell, it may reasonably be assumed that the variation in the heterocaryon characters resulted from a varying proportion of the parental nuclei.

ACKNOWLEDGMENTS

The author wishes to express his appreciation to Dr. Y. Tanaka and Dr. M. Tsujita for their interest and advice during this investigation.

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EXPLANATION OF PLATES

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- Fig. 1. Aspergillus candidus. Segregation of a colony obtained from the heterocaryon into two patches, heterocaryotic and homocaryotic.
- Fig. 2. Aspergillus awamori. Hyphal anastomosis between two hyphae derived from No. 5-9 and No. N-19.
- Fig. 3. Aspergillus awamori. Left, No. 5-9; right, No. N-19; bottom, their heterocaryon.
- Fig. 4. Penicillium chrysogenum. Left, U. V. mutant, UY-3S; right, U. V. mutant, UW-1S. In the middle two tubes development of the heterocarya with green conidia as in the wild type.
- Fig. 5. Penicillium chrysogenum. Morphological characters of colonies obtained from the heterocaryon between the two U.V. mutants showing a continuous variation between the parents.

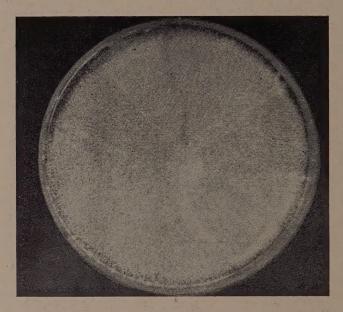


FIG. 1



FIG. 2

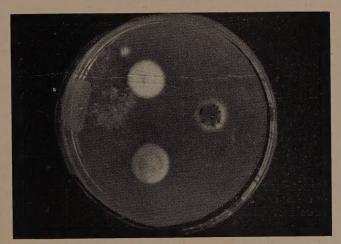


FIG. 3

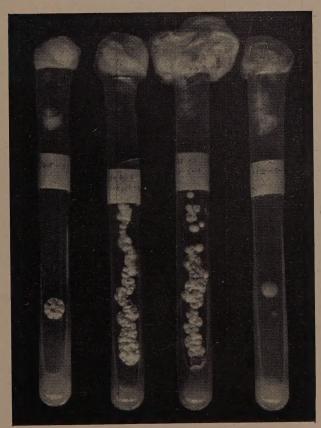
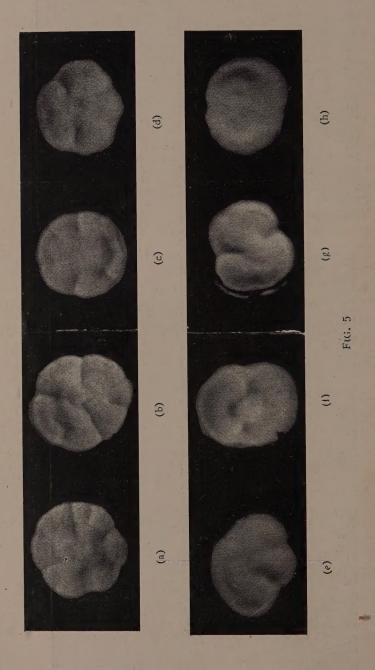


FIG. 4



A NEW FUNGUS, MONOCILLIUM INDICUM GEN. ET SP. NOV., FROM SOIL*

S. B. SAKSENA

(Accepted for Publication, July 12, 1955)

This genus was isolated from a sample of grassland soil of Patharia village near Sagar. The fungus first appeared in a plate of Waksman's agar and was cultivated on Czapek's agar for the purpose of recording observations and measurements.

General Structure: The remarkable feature of the genus is the pattern of conidiophores and conidia. The conidiophores are simple, unbranched and possess a characteristic shape (Fig. 1—C, Pl. 1) in having a long pedicel surmounted by a typical phialide which produces a long chain of spores (Fig. 1—A & B, Pl. I). The spores are usually ovate with one end rounded and the other pointed (Fig. 1—D). In the arrangement of the spores the pointed end is directed towards the proximal side (Fig. 1—B).

Since there is no development of colour, the fungus clearly belongs to the family Moniliaceae. The chain of spores formed in basipetal succession would suggest a kinship with such genera as *Penicillium* and *Paecilomyces* but the form and structure of conidiophore is clearly different from any of the known genera. The fungus is, therefore, described as a new genus. It is named *Monocillium* after its simple unbranched conidiophore and the species which is the only one known at present, is named *Monocillium indicum* after the country.

Monocillium gen. nov.

Coloniae incolorae, lento crescentes; conidiophori simplices, non ramosi, septati, constantes longo pediculo cui insidet unica phialis typica, quae producit catenan conidiorum. Conidiorum catenae longae. Conidia ovata vel elliptica, laevia.

Colonies colourless, slow growing, conidiophores simple, unbranched, septate, consisting of a long pedicel surmounted by a single typical phialide producing a chain of conidia. Conidial chains long. Conidia ovate to elliptical, smooth.

Monocillium indicum sp. nov.

Coloniae lentissime crescunt in "Czapek agar", diametrum attingentes 1-2 cm. post 10 dies in temperie normali cubiculi, incolorae, tenues, demissae. Hyphae vegetabiles submersae, ramosae, septatae, incolorae, ca. 1.5μ . crassae. Hyphae aereae efformant

^{*}Part of the thesis approved for the Degree of Doctor of Philosophy by the University of Saugar.



PLATE I

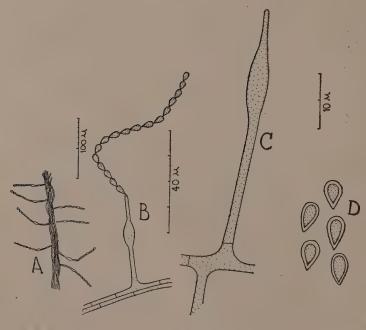


FIG. 1

flocculentiam tenuem, saepe monstrant structuras funiculares delicatulas, sparse ramosae, septatae, ca. 1.5 μ . crassae. Conidiophori insidentes hyphis aereis, simplices, non ramosi, septati, $35-50\mu$. longi, constantes pediculo longo superato unica phialide typica quae producit catenam conidiorum; pediculus $20-30\times1.5\mu$, phialis $20-25\times2.1-3.5\mu$; conidiorum catenae usque ad 130μ longae. Conidia ovata vel elliptica, pallide luteobrunnea, apice altero rotundato, altero vero acuto, laevia, crassis parietibus praedita, $5-7\times3-4\mu$.

Colonies on Czapek's agar growing extremely slowly, attaining a diameter of about 1–2 cm. in 10 days at room temperature, colourless, thin, slow growing, vegetative hyphae submerged, branched, septate, colourless, about 1.5μ thick. Aerial hyphae forming a thin flocculence, often showing rather delicate ropy structures, sparsely branched, septate, about 1.5μ thick. Conidiophores borne on the aerial hyphae, simple, unbranched, septate, $35-50\mu$ long, consisting of a long pedicel surmounted by a single typical phialide producing a chain of conidia; pedicel $20-30\mu\times1.5\mu$, phialide $20-25\mu\times2.1-3.5\mu$; conidial chains upto 130μ long. Conidia ovate to elliptical, light yellowish brown, with one end rounded and the other pointed, smooth, thick walled, $5-7\times3-4\mu$.

Cultural characteristics: The fungus was grown on several culture media with a view of ascertaining its behaviour and range of variability. On malt agar and potato-dextrose-agar the growth was faster and colony was thicker than on Czapek's agar. There was no significant difference in the ranges of measurements. In case of soil extract agar the growth was slow and the spores tended to accumulate in the form of balls at the apices; other characters remained the same.

The type culture is being deposited with the Indian type culture collections, Division of Mycology of the Indian Agriculture Res. Instt., New Delhi.

Spore germination: Spore germination was tried in pea decoction. The spores readily germinated in about 6-8 hours giving out a single germ tube.

Affinities: It is not infrequent to find single phialides developing in the genus Paecilomyces and in diminutive forms of some Penicillia (Raper and Thom, 1949), but in this genus the unbranched single conidiophere is a fixed rule. Secondly the long pedicel seen below each phialide is a characteristic structure not found any where else (Clements and Shear, 1931; Gilman, 1945). The basipetal chains of conidia will undoubtedly place it near the above mentioned two genera,

SUMMARY

A new genus of Moniliaceae is described. The fungus resembles the genus *Paecilomyces* and *Penicillium* in producing basipetal chains of spores on phialides. It is characterised in having simple, unbranched conidiophores consisting of a single phialide which is supported on

a long pedicel. The genus is named *Monocillium* and the species *Monocillium indicum*. The cultural characteristics on several media are given. Spore germination was also studied.

ACKNOWLEDGMENTS

The writer wishes to express his grateful thanks to Dr. R. K. Saksena for his kindly guidance and advice and to Dr. R. Misra for the facilities and encouragement. He is very much obliged to Dr. Charles Thom for kindly going through the description and the figures of the fungus and to Prof. Fr. H. Santapau for the Latin diagnosis.

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EXPLANATION OF FIGURE—1

A—Habit, showing the development of conidiophores × 150.

B—A single conidiophore bearing chain of conidia \times 600.

C—A single conidiophore magnified, showing a long pedicel and the terminal phialide \times 1050.

D-Conidia ovate with one end round and the other pointed × 1050.

EXPLANATION OF PLATE-I

Showing conidiophores which are unbranched, pedicellate structures bearing chains of conidia × 400.

FURTHER STUDIES ON ASPERGILLUS BLIGHT OF GROUNDNUT SEEDLINGS: ITS OCCURRENCE AND CONTROL

K. G. Nema, A. C. Jain and R. P. Asthana.

(Accepted for publication, July 14, 1955)

INTRODUCTION

A seedling disease of groundnut (Arachis hypogaea L.) was observed at Nagpur in July 1950. Jain and Nema (1952) determined Aspergillus niger van Tiegh. as the cause of the disease and worked out the morphology of the pathogen. Earlier Jochem (1926) had reported a seedling disease of groundnut from Sumatra due to A. niger which was later on identified as A. pulverulentus (Mc Alpine) Thom by Boedijn (1928). Morwood (1945, 1946) from Queensland, Blackie (1947) from Fiji and Wallace (1948) from Tanganyika described a 'Crown rot' disease of groundnut seedlings caused by Aspergillus sp. Later Gibson (1953) from South Africa has reported the crown-rot of groundnut seedlings identifying the pathogen as Aspergillus niger van Tiegh. Morwood (1953) also found A. niger as the cause of crown-rot in Queensland,

SYMPTOMS

The disease appears in two phases *i.e.* the pre-emergence phase and the post-emergence phase. In the former, the parasite causes a rotting of the germinating seeds and kills the seedlings before their emergence from the soil, (Plate I, Fig. 1), while in the latter, the disease is characterised by wilting and death of the seedlings accompanied by a rotting of the hypocotyl region (Plate I, Fig. 2). In the latter phase, a circular light brown spot appears on the cotyledon in the initial stage. This discoloured area rapidly loses its normal hardness due to rotting. The infection spreads on to the hypocotyl and stem region. The vascular tissues are shredded, with the result that the affected plants collapse. Grayish white mycelium, with black fructification of the pathogen, appears on the surface of the affected parts. (Plate I, Fig. 3). These symptoms are evident under humid conditions but under dry conditions the lesions remain localised. Secondary infection from plant to plant has not been observed.

These observations suggest the possibility of the perpetuation of the disease through seed or soil. Seeds carrying infection give rise to the diseased cotyledons which in turn infect the hypocotyl or stem of the seedlings. The pathogen present in the soil either infects the cotyledons or the hypocotyls direct. The above observations are similar to those obtained by Gibson (1953) in East Africa. In order to obtain the reliable data on the reccurrence of the disease a number of experiments were made.

Aspergillus Blight of Groundnut Seedlings



F1G. 1



F1G. 3



FIG. 2

EXPERIMENTAL

Ten-day old pure cultures of Aspergillus niger, multiplied on Czapek agar and AK 10 variety of groundnut were used throughout the experiments unless otherwise stated.

Surface sterilised seeds were inoculated with cultures of A. niger and were sown in sterilised soil in pots. All such pots were kept under bell-jars to maintain sufficient humidity. The results obtained are given in Table 1.

The data reveal that the presence of mycelium is essential for infection of uninjured seeds. Conidia alone are capable of infecting the seeds only when testa is broken. Even in cases of injured seeds the combination of mycelium with conidia caused 100% infection, whereas conidia alone produced only 25% infection. If the injury is extended to the cotyledons, the infection is so rapid that the seedlings die before emergence. The mycelial inoculum caused 100% and condial inoculum about 33% pre-emergence blight in case of injured cotyledons.

The effect of soil inoculation on the infection of seeds with testa intact and broken was further studied. Surface sterilised seeds with intact and broken testa were sown in sterilised soil which was inoculated with the pathogen a week before sowing. Regular control was maintained. The disease appeared only in inoculated soil. The results are tabulated in Table 2.

Forty healthy seedlings each raised from seeds with unbroken testa in the inoculated and uninoculated soil were slightly injured just below the soil surface, when they were 21 days old. They were kept under observation to see whether infection could be caused directly to the collar region by the inoculum in the soil through wounds. None of the seedlings raised in the uninoculated soil were infected while 20 percent of the seedlings raised in the artificially infected soil showed typical lesions of the disease after seven days of the injury.

The above results indicate that the seedlings are more susceptible to the pathogen in the soil when the seeds or the seedlings are injured. Morwood (1945, 1946) has reported similar findings and has stated that the pathogen A. niger enters the host through wound on the seed coat (testa) or the stem. From the above, it is concluded that for healthy growth seeds or seedlings should be in sound condition at the time of sowing or in growing stage.

A number of experiments were carried out to observe the effect of seed treatments with fungicides on the incidence of the disease. Seeds with broken testa were inoculated with the pathogen and then such seeds were treated with the recommended doses of the requisite fungicides. The observations are recorded in Table 3 and indicate that seed dressing with Ceresan, Agrosan GN and Fernasan A, are most effective in pre-emergence stage while Ceresan and Agrosan GN, also appreciably reduce the incidence of post-emergence killing.

ABLE 1

Inoculation of seeds and percentage infection of Aspergillus Blight.

Percentage infection after 10 days of inoculation	28.5	100.00	100.00*	Nil Nil Nil *Seedlings died before emergence.
Method of Inoculation	Placed on seed-surface with intact testa.	Placed on seed surface with broken testa.	Placed on seed surface with seed cotyledons injured.	 (a) Seeds with intact testa. (b) Seeds with broken testa. (c) Seeds with injured seed cotyledon.
Treatments	Mycelium + Few spores Spores suspension in sterile water.	Mycelium + Few spores. Spores suspension in sterile water.	Mycelium + Few spores. Spores suspension in sterile water.	Control (No inoculum)
S. No.	1. (a)	2. (a) (b)	3. (a) (b)	4

Incidence of the disease in infected soil. TABLE 2

				Nu	mber of	Diseased	Number of Diseased Seedlings	S	Total	
Š,Z	Treatments	No. of seeds	of sow-	Pre-emergence	gence	Pos	Post emergence.	nce.	No. of diseas- ed	Percent- age in- fection
		sown.	14-8-51	21-8-51		23-8-51	23-8-51 29-8-51 31-8-51	31-8-51	seed- lings.	
ri .	Control—No pathogen Seeds with									
	(a) Intact Testa. (b) Broken Testa.	40		ALL S	EEDLIN	VGS REN	AAINED	ALL SEEDLINGS REMAINED HEALTHY	ж	
72	Soil with pathogen. Seeds with									
	(a) Intact Testa.	80		∞		1	1	I	∞	10.00
	(b) Broken Testa.	98		56		4	11	Ŋ	46	57.50

TABLE 3

Effect of seed treatment on the incidence of Aspergillus Blight.

		Number of	Number of Seedlings Affected	Affected	Perc	Percentage Infection	ction
Treatments	No. of seeds inoculated & sown	Pre- emergence emergence killing killing	Post- emergence killing	Total No. of seedlings affected	Pre- emergence killing	Post- emergence killing on the basis of No. of seeds sown	Total No. of killing on the basis of No. of seeds sown
1. Control 2. Copper Carbonate 3. Aagrano 4. Tritisan 5. Tillex 6. Fernasan A 7. Agrosan GN 8. Ceresan	<i>2</i> 222222	4.08801	9819442	98841 98421 984	60.71 14.28 10.71 3.57 0.00 0.00	10.71 53.57 28.55 10.71 7.14 7.14 3.57	71.42 57.14 42.83 21.42 10.71 7.14 3.57

Active ingredients:—AAGRANO—(3-ethoxypropyl) mercury bromide; TRITISAN—pentachloronitrobenzene; TILLEX—15% Ethyle mercury chloride: FERNASAN A—bis (dimethylthiocarbamoyl) disulphide; AGROSAN GN—tolylmercury acetate; CERESAN—N-(ethylmercuri) p-toluenesulfonanilide.

Varietal Susceptibility of Groundnut to Aspergillus Blight. TABLE 4

Percentage infection	Post- emer- gence killing	3.75 9.51 16.13 63.16 88.71 63.16 84.21 84.21 84.20 84.44 83.33 29.63
Perc	Pre- emer- gence killing	93.75 81.95 72.58 21.05 46.00 38.89 53.70
affected	Total No. of affected seedlings	\$470.844.4 \$40.844.4
Number of seedlings affected	Post- emer- gence	16 10 13 13 14 16 16
Number	Pre- emer- gence	\$2\$228 \$2
No. of	Seeds inocula- ted and sown	. 27.20.20.80
	Variety	1. AK 10 2. Spanish peanut 3. E. C. 1733 4. AK 8-11 5. AK 12-24 6. Small Japan 7. Kopargaon

The results thus suggest the beneficial fungicidal action of seed dressing when seed-surface is contaminated with A. niger.

In another experiment 7 varieties of groundnut, viz., AK 10, AK 12-24, AK 8-11, Small Japan, Spanish Peanut, E. C. 1733, and Kopergaon; kindly supplied by the Economic Botanist to Government, Madhya Pradesh, were tested for their susceptibility to Aspergillus Blight. Surface sterilised seeds with broken testa were inoculated with pure cultures of A. niger, and sown in 11'×6' plots. The results are tabulated in table 4 and indicate that all the varieties tested and predominently used in the State of Madhya Pradesh are susceptible to the disease. As such no definite variety can be recommended against the Aspergillus Blight disease.

SUMMARY

- 1. Experiments on the possibilities of the occurrence of Aspergillus blight of groundnut seedlings caused by Aspergillus niger van Tiegh have been carried out.
- 2. Inoculum for hypocotyl infection may be provided by the pathogen when present either on the seed or in the soil.
- 3. Seed or stem injury is an important factor for inducing the disease.
- 4. The presence of mycelium is essential to induce infection in seeds with unbroken testa.
- 5. Seed treatment with fungicides such as Ceresan, Agrosan GN and Fernasan A have been found beneficial at pre-emergence stage and with Ceresan at post-emergence stage.
- 6. All varieties of groundnut prevalent in Madhya Pradesh have been found equally susceptible to the disease.

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EXPLANATION OF PLATE

- Fig. 1 Pre-emergence phase—showing affected seedlings before their emergence out of the soil.
- Fig. 2 Post-emergence phase—showing wilting of plants and rotting of cotyledons and hypocotyl regions.
- Fig. 3 Affected hypocotyl region showing sporulation of Aspergillus niger.

PREVALENCE OF PHYSIOLOGIC RACES OF WHEAT AND BARLEY RUSTS IN INDIA

R. S. Vasudeva, R. Prasada, V. C. Lele, L. M. Joshi and D. Kak

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Wheat is the second most important cereal crop in India with nearly 24 million acres under cultivation. An important factor which is responsible for considerable reduction in yield is the attack of rusts. All the three rusts, viz. black, brown and yellow, caused by Puccinia graminis tritici (Pers.) Erikss. & Henn., Puccinia triticina Erikss. and Puccinia glumarum (Schm.) Erikss. & Henn. respectively, are common on wheat. Puccinia graminis and Puccinia triticina are prevalent in almost all the wheat-growing regions but Puccinia glumarum has a restricted distribution, being common only in the Northern parts of the country, though it is not altogether absent from South India. In the Northern parts of the country, which comprise the main wheat growing States, all the 3 rusts appear year after year causing considerable damage to the crops, particularly in the years of serious epiphytotics.

The study of physiologic races of rusts for implementing breeding programme to control rust epidemics is of fundamental importance. So far no connected account is available to show the occurrence and distribution of races in different regions of the country. With this object in view information so far available is recorded in this paper. In order to provide as complete a picture as the available data would permit, information has been freely drawn from the work published earlier by Mehta (1940), Prasada & Lele (1952) and Vasudeva et al (1953).

The methods of collection of rust samples and their study was the same as described by Mehta (1940). Differential hosts originally selected by Stakman and Levine (1922) were used for the study of black rust, those selected by Johnson and Mains (1932) for brown rust and those selected by Gassner and Straib (1932 and 1934) for yellow rust.

During a period of twenty years *i.e.* from 1932-1952, 3290 rust collections were analysed and eleven races of *Puccinia graminis tritici*, eight races of *P. triticina* and ten of *P. glumarum*, have been recorded in this country. Mehta (1940) reported the occurrence of races 15, 21, 24, 40, 42 and 75 of black rust, of races 10, 20, 63, 106, 107 and 108 of brown rust and of races 13, 19, 20, 31, A*, D*, E*, and F* of yellow rust. Prasada and Lele (1952) reported races 34, 117** and 194 of

^{*}These races were identified for the first time in India and have not been assigned international numbers.

^{**}Race 117 is identical with the race recorded by Uppal and Gokhale (1947) and which they stated is closely allied to race 119. This race has now been assigned Race No. 117 in consulation with Dr. E. C. Stakman.

black rust, race 11 and 26 of brown rust and races G^* and H^* of yellow rust. Gokhale and Patil (1952) identified race 122 from Bombay State. One more race of black rust, which is closely allied to race 72 has also been recorded by Vasudeva, Lele and Misra (1953). In addition to these races two biotypes of race 42 (Uppal and Gokhale, 1947) and one of race 15 (Gokhale, 1950) of black rust have also been found. Biotypes 42-B and 15-C have been found to be highly virulent infecting a large number of varieties resistant to other races.

Experimental Results:—Even though the number of samples analysed for occurrence of physiologic races in the country in the last 20 years has been rather small, it is possible to draw certain general conclusions regarding population shifts from the studies of the annual prevalence and distribution of these races. In tables 1, 2 and 3 the occurrence of physiologic races in different States of India and Nepal has been shown. It will be found that different races are more or less uniformly distributed throughout the country and there does not appear to be any regional specificity obviously on account of absence of any natural barriers. Another important factor responsible for the uniform distribution of races is the absence of extensive cultivation of rust resistant varieties and continuous cultivation of particular set of varieties in different regions.

Besides the study of rust samples from wheat and barley crops, samples from some grasses have also been studied and the results of analysis are set out in table 4.

From this data it is clear that there is no zonal distribution of races and the common races of all the rusts are prevalent in almost all parts of the country where a particular rust appears. Some of the races mentioned here are by no means restricted to wheat and barley and have also been picked up from grasses and rye.

In tables 5, 6 and 7 information regarding frequency of races, calculated on the basis of the number of times a race was isolated in relation to the total number of times all races were identified during the year together with the total number of samples analysed during the year, is presented. The word "isolate" signifies a race picked up from a single collection and its use has been necessitated by the fact that in some cases more than one race is isolated from a single collection. When two races are present in the same collection they produce two kinds of uredia or two different infection-types on the same variety (differential-host) and can be identified by making suitable isolations from them and each is designated as an "isolate".

^{*}These races were identified for the first time in India and have not been assigned international numbers.

TABLE Occurrence of physiologic races of Puccinia

		KASHM	IR	(includi	PUNJA ing H. PE	
Year	No. of Sta- tions	No of Samp- les	Races	No. of Sta- tions	No. of Samp- les	Races
1932-33	angum.	_	_	18	30	15, 40, 42, 75
1933-34	1	1	15, 40	\{ 11	18	15, 21, 24, 40, 42, 75
1934-35	·2	2	15, 42	(1 {18	1 * 19	15 15, 24, 40, 42, 75
1935-36	{ 10 2	10 2*	15, 40, 42 15	\ \{ 21 \ \ 4 \ 4	4* 26 4*	15, 40, 42 15, 40, 42 15, 42
1936-37	{ 9 { 2	9 2*	15, 40, 42 15, 42	{ 14 7 3	18 3*	15, 40, 42 42
1937-38	4	4	15, 40, 42		19 4* .	15, 40, 42 15, 40, 42
1938-39	1	1 .	15	{ 19 { 4 { 11 { 2	11 2*	40, 42 40
1939-40	4	6	15, 40	5	6	15, 40, 42
1940-41	1	1 .	40	3	6 .	15, 40, 42
1941-42	функци	· -		1	4 :	15, 40, 42
1942-43		- .	all and the same of the same o	§ 3	3 1*	15, 42 42
1943-44			-	\ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \	2 1*	15, 40, 42 15
1944-45	1	1.	15, 42	3	4	15, 40, 42
1945-46		_			-	
1946-47	drauga	almora				-
1947-48		_	_	_		_
1948-49 1949-50	-	_		, $\frac{\cdot}{2}$	4	21, 40, 42
1950-51		****	,	.3	3	21, 34, 42
1951-52	_			$\left\{\begin{array}{c} 7 \\ 2 \end{array}\right\}$	14 2*	21, 24, 34, 40, 42 21, 34, 40
Total No. of wheat samples Total No. of barley samples		35 4			187 22	-

Note:—‡ Altogether two samples, one from 1941-42 and the other from 1950-51 races 21 and 42 only.
† Altogether three samples, one each from 1936-37, 1937-38 and 1951-52 and 42 and the last race 21.
* Indicates barley samples.

1 graminis tritici in different States.

DELHI	UTTAR PRADESH	BIHAR AND ORISSA	BENGAL AND ASSAM	
No. of No. of Sta- Samp- Races tions les	No. of No. of Sta- Samp- Races tions les	No. of No. of Sta- Samp-Races tions les	No. of No. of Sta- Samp-Races tions les	
1 1 15, 24, 42, 75 1 1 15, 75	18 20 15, 40, 42, 75 20 22 15, 40, 75	4 8 15, 40, 42, 75 { 3 4 15, 40, 75 { 1 1* 15	2 2 15, 40, 42	
\begin{cases} \begin{cases} 1 & 1 & 42 \\ 1 & 1* & 42 \\ 1 & 1 & 40 \\ 1 & 1* & 15, 42 \\ 1 & 1 & 15, 40 \end{cases} \end{cases}	{12 19 15, 40, 42, 75 1 1* 42 {10 14 15, 40, 42, 75 1 1* 42 {11 13 15, 40, 42}	3 6 . 15, 40, 42, 75 75 1 2* 15, 40 1 1 40 1 1* 15 1 2 15, 42	2 2 40, 42	
1 1 15, 40 {1 1 15, 42 1 1* 40 1 1 15	3 3* 15, 40, 42 10 10 15, 40, 42 4 4* 15, 42 5 6 15, 40, 42	\$\begin{cases} 3 & 3 & 15, 40, 42 \\ 2 & 2* & 15, 42 \\ 3 & 3 & 15, 40, 42 \\ 1 & 1* & 42 \\ 2 & 8 & 15, 34, 40, \end{cases}\$	{3 3† 15, 40, 42 1 1* 15, 42 1 1 40, 42	
$ \begin{cases} 1 & 1 & 40,42 \\ 2 & 1* & 15,34 \\ 1 & 4 & 15,24 \\ 40,42 \\ 1 & 1* & 40,42 \end{cases} $	{ 10 16 15, 40, 42	1 3 34,40 2 2‡ 15,42		
1 1* 40, 42 1 2 42 1 1* 15, 42 1 1 40 1 1* 15, 42 1 1 21 1 1* 21, 40, 42 1 1 42 1 1 1* 15 1 2 21, 40, 42	{ 10 10 15, 40, 42 5 5* 15, 40, 42 3 5 15, 40, 42 2 2 21, 40	\$\begin{cases} 4 & 9 & 15, 40, 42 \\ 1 & 1* & 15, 40, 42 \\ 1 & 1 & 15 \\ 1 & 1* & 15 \\ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \		
	$ \begin{cases} 1 & 1 & 21 \\ 16 & 16 & 15, 21, 34, \\ & 40, 42, 117 \\ 3 & 3* & 21, 40, 42, \\ & 194 \end{cases} $	\[\begin{pmatrix} 1 & 3 & 15, 21, 42 \\ 1 & 1* & 15 \\ 2 & 2 & 21, 40, 42 \\ \end{pmatrix} \]		
\$\begin{cases} \begin{cases} 1 & 4 & 21, 42 \\ 1 & 1* & 21 \\ 1 & 4 & 21, 40, 42 \\ 1 & 1 & 21 \end{cases}\$\$	7 8 21, 42 7 9 21, 40 3 5 21, 40 3 4 21, 24 & 42	1 1 21 1 7 21, 40, 42 1 2* 21 1 2 5‡ 21, 42 2 3* 21, 34 3 6 21, 34	 _ 1 1 21,42	
3 5 21, 24, 40 1 1 40	$ \begin{cases} 21 & 37 & 21, 34, 40, \\ 42 & 2^* & 34, 40 \end{cases} $	2 3 21, 42 2 3* 21, 34 3 6 21, 34	2 2† 21,34	
34 10	246 34	74 15	13 1	

crops, were received from Orissa. The former yielded race 42 and the latter crops, were received from Assam. The first one yielded race 42, second race 40

TABLE

						IABLE
	MADE	IYA PRA BERA	ADESH AND AR		DHYA I	BHARAT DATIA)
Year		No. of samples	Races		No. of samples	Races
1932-33	1	1 .	15	1	1	40
1933-34	, 1	1	15, 42	2	2	15, 40
1934-35	`4	. 7	15, 42	2	6	15, 40, 42
. 1935-36	. 4	. 5	15, 40, 42	$\left\{\begin{array}{c} 1\\1\end{array}\right.$	5 2*	15, 40, 42, 75 40, 42
1936-37	4	10	40, 42	1	4	42
1937-38	4	6	15, 40, 42			_
1938-39	1	1 .	42			
1939-40	2	6	15, 40, 42		tente.	
1940-41	3	9	15, 40, 42	1	2	40
1941-42	{ 14 1	17 1* .	15, 34, 42 15	$\begin{cases} 1 \\ 1 \end{cases}$	2 1*	34, 42 15, 34, 42
1942-43	14	14	15, 40, 42	4	4	15, 40, 42
1943-44	{17 { I	20	15, 40, 42 40	1	1	40
1944-45	- 13	15	15, 21, 40, 42, 117, 194	1	. 1	21, 40, 42
1945-46	1	1	42	_		-
1946-47	27	31	15, 21, 34, 40, 42, 117	· 4	5	21, 42, 117
1947-48	28	37	15, 21, 42, 117	_	- Common	-
1948-49	{15 1	16 2*	15, 21, 40, 42, 117 40, 42		_	
1949-50	16	22	21, 40, 42	1	1	21, 42
1950-51	3	3	21, 40, 42		-	_
·1951-52	$\begin{cases} 4\\1 \end{cases}$	4 1*	21, 34, 40, 42 21, 42	2	2	21, 34
Total no. of wheat samples Total no. of barley samples		226 5			36 3	BAV

1 (Continued)

	RAJAST		-	ВОМВ	AY	: S.	AURAS	HTRA
	No. of samples	Races	No. of stations	No. of samples	Races	No. of stations	No. of samples	Races
1	1	40, 42	11	33	15, 40, 42, 75	2	3	15, 40, 42
1	1	42	5	7	15, 40, 42	2	2	15, 40
4	5	24, 42	10	21	15, 24, 40, 42	.3	6	15, 42
_			{13 1	22 1*	15, 40, 42 40	3	6	40, 42
{ 2 1	4 1*	42	{ 25 2	34 2*	15, 40, 42 42	13	15	15, 42
-	<u>:_</u>		2	3	15, 42	6	6	15, 40, 42
Minguel		-		portes	-	4	4	15, 40, 42
Million			.3	3	40, 42	1	2	15, 40, 42
	_	Standing	6	. 6	15, 40, 42	3	6	15, 34, 40, 42
-	_		2	3	34, 42	· 1	3	40
-		_	2	4	40, 42	-		
1	2	15, 40, 42	14	16	15, 40, 42	4.	6	15, 40, 42
		BBase	17	21	15, 21, 40, 42, 117, 194	5	8	15, 21, 40, 42
ground		_		-	-	7	1	15, 42
	*****		10	12	15, 21, 40, 42, 117	4	5	21, 42, 117
		Windows	11	14	15, 21, 40, 42	11	15 .	15, 21, 42, 117
-	-	_	8	10	15, 21, 34, 40, 42	5	6 .	21, 40, 42
-		_	7	10	21, 34, 40, 42		_	_
-	-		5	6	21, 34, 42	-		_
	_	_	4 .	4	21, 42	2	2	21
				200		_	96	
	13			229			90	
	1			3				

TABLE

						TABLE		
Year		внор.	AL		MADRAS			
Iear	No. of stations	No. of samples	Races	No, of stations	No. of samples	Races		
1932-33				1	3	15, 40		
1933-34			-	8	9	15, 40, 42		
1934–35	<u>.</u>		-	1	1	42		
. 1935-36	,			{ 5 1	6 1*	15, 40, 42 42		
1936 37	.1	1 .	42	{ 10 1	20 1*	15, 42 42		
1937 38	-		ethelised.	{ 17 1	. 1*	15, 40, 42 40, 42		
1938-39		· · ·		{ 7· 1	7 1*	15, 40, 42 40		
1939-40			Minutes	{ 9 { 1	10 1*	15, 40, 42 42		
1940-41	-		-0	· 2	2	15, 40		
1941-42	_	~		{ 1 1	1 1*	34, 42 42		
1942-43			·	1	2	21, 42		
1943-44				. 4	4	15, 40		
1944-45	-	 .	*****	4	5	21, 42		
1945-46		,	-		· ·	_		
1946-47				4	5	21, 42, 117		
1947-48	2	2	21, 42	1	1	21		
1948-49	_	-	B	3	3	21, 42		
1949-50	-	Promp		-		. —		
1950-51	3	4	21, 42	1	1	21		
• 1951-52	1	1	21	{ 2 2	2 2*	21, 34, 42 21		
Total No. of wheat samples Total No. of barley samples		8 —			104	mile		

NOTE:—Altogether three samples of wheat received from Travancore-Cochin One sample of barley received from Travancore-Cochin in (1939-40) Altogether five samples of wheat received from Afghanistan in

Total No. of samples:-

1—(continued)

HYDERABAD				MYSORE				NEPAL			
No. of stations			No. of stations	No. of samples	5	Races	No. of stations	No. of samples	Races		
3	5	15, 40, 42	1	1	40,	42	_	_			
2	3	24, 40, 42	3	3	15,	42		,	_		
2	10	40, 42, 75	2	10	15,	40, 42	1	· 1	75		
	1	42	3	6	42		.7	7	15, 42		
3	7	15, 40, 42	4	4	40,	42	·6	6	15, 40, 42, 75		
2	3	15, 42	3	3	42		6 -	6	15, 40, 42		
1	1	42	2	2	42		6	6	15, 40, 4.		
1 '	2	15, 42	4	4	15,	40	.	-	_		
2	3.	15, 42	2	2	15,	40		_	_		
-	-		1	3	42		_	, 	_		
2	3	15, 40, 42	2	. 3	24,	42	_		_		
3	3	15, 40, 42	4	4	15,	40, 42			Applica		
4	5	21, 40, 42	{ 5 1	· 5 1*	21, 40	42		-	~		
	_	_		(Massie			-	passer.			
		_	2	2	15,	42	_	_	-		
3	3	21, 42	2	2	21,	42	-				
5	5	21, 42	1	1	42		-	-	-		
6	11	21, 40, 42	1	1	21,	, 42	-		-		
2	5	21, 24, 34, 40, 42	4	4	21,	, 42		-	-		
	_		_		_		_				
	70			60				26			
	Markey			1							

in (1950-51) yielded races 21 & 40. yielded race 42. (1938-39) yielded races 15 and 40. Wheat=1465 Barley=108.

TABLE Occurrence of physiologic races of

	e of physiologic races of						
Year		KASH	MIR	PUNJAB (including H. PRADESH)			
1 car	No of No. of stations samples		Races	No. of No. of stations samples		Races	
1932-33	1	1 .	10, 63	20	29	10, 20, 63	
1933-34	*******			9	10	10, 20, 63	
1934-35	-	_	_	16	17	10, 20, 63, 106, 107, 108	
1935-36	8	8	20, 63, 107,	14	22	10, 20, 63, 107	
1936-37	.1	1	63	16	19	10, 20, 63	
1937-38	1	1	20	16	19	20, 63	
1938-39		-		9	9	20, 63	
1939-40		_	-	_			
1940-41	<u></u>	·		2 .	3	20, 63	
1941-42	1	1	63	2	2	20, 63	
1942-43		-	-	. 4	4	10, 20, 63	
1943-44				1 .	1	10	
1944-45		-		2 .	2	20	
1945-46	<u>.</u>	1		,	-		
· 1946-47	<u> </u>	-	-	-	-	<u> </u>	
1947-48		asinor					
1948-49			_			_	
1949-50			-	1	1 -	20, 63	
1950-51	_			3	5	11, 20, 63 106, 108	
19 5 1-52		~~~	_	5	11	20, 26, 63, 106, 108	
Total No. of wheat samples		12			154		

-2 Puccinia triticina in different States

	DEL	HI	UTT		RADESH	SH BIHAR AND ORISS		
No. of stations	No. of sample		No. of stations			No. of stations		Races
1	1	10, 63	20	28	10, 20, 63	3	5	10, 63
1	°1	63	22	22	10, 20, 63, 106	6	, 6	20, 63
1	1	20	22	26	20, 63, 108	3	6	10, 20, 63
1	1	20, 63	18	26	20, 63, 107	11	14	20, 63, 107
1	. 1	63	24	31	10, 20, 63	13	17	10, 20, 63
1	4	20, 63	14	15	20, 63	. 4	6	20, 63
1	1	20	11	13,	20, 63, 107	3	3	20
1	1	63	14	18	10, 20, 63, 107	2	8	10, 20, 63
1	1	20	21	23	10, 20, 63, 108	2 ·	4	10, 20, 63
1	1	63 .	19	2 5	20, 63	1	3	20, 63
1	. 2	10, 20, 63	16	18	10, 20, 63	4	6	20, 63
1	1	20, 63	6	8	10, 20, 63	. 1	1	63
1	2	20	2	2	20 ·	_		
1	1	107	1	. 1	63, 107	1	1	107
1	1	11	12	13	20, 26, 63, 106, 108	1	1	20
1	5	19, 20, 26, 63, 108	7	8	20, 26, 106	1	2	20, 26
1	2	10, 20, 63	7	8	10, 11, 20, 63	1	1	20, 63
1	1	63, 108	3	8	20, 26, 63, 108	1 .	5	11, 20, 63, 106
4	4	20, 26, 63; 108	2	2	20	4	4*	20, 63, 106, 108
1	4	20, 106	18	27	10, 11, 20, 26, 63, 106	2	3	20, 63, 108
	36			322			96	

^{*} Altogether two samples were received from Orissa and they yielded races 20, 106 and 108.

TABLE

						IABLE	
	BEN	GAL AN	D ASSAM	MADHYA PRADESH AND BERAR			
Year		No. of s samples	Races		No. of as samples	Races	
1932-33	2	2	10, 63	_		_	
1933-34	. 2	· 2 ·	20, 63	2	2	10, 20	
1934-35	2 ·	. 3	20, 63	4	6	20, 63	
1935-36	-1	2	63	-	_	_	
1936-37	9	10†	10, 20,63		_		
1937-38	7	9†	20, 63	-	_	_	
1938-39	6	6†	20, 63	3	3	20, 63	
1939-40	. ~~	_	garant.				
1940-41	2	2	63	2	4	20	
1941-42	—	-	-	6	7	20, 63	
1942-43	-		-	2	4	10, 63	
1943-44			_	2	2	20, 63	
1944-45	ARRIV		.	3	3	10, 63, 107	
1945-46	_	-	-		*******		
1946-47	· -	. 	-	16	17	10, 11, 20, 26, 63, 107	
1947-48	-		Millerman	1	2	20, 26	
. 1948-49	 .		***************************************	11	14	10, 11, 20, 63, 108	
1949-50				2	3	10, 11, 20, 63	
1950-51	4	9 .	10, 11, 20, 63, 106, 108	—		-	
1951-52	1	1	11, 20	3	4,	20, 63	
Total No. of wheat samples		46			71		

[†] Altogether eight samples were received from Assam. One sample from and one sample from 1938-39 crop yielded race 20 only.

2—(continued)

MAI (inc	DHYA cluding	F	RAJAS	THAI	N	BOMBAY			
No. of stations	No. or	f Races	No. of stations	No. or	f es	Races	No. of stations	No. o	f es Races
			-		-		_		_
2	2	20	2	2	10, 6		4	4	20, 63
1	1	10, 20	5	6	20, 1	.07	2	. 2	10, 20, 63
e-many				_				_	
We wrong	enna.		1	3	20, 6	53		Minima	
					_				
-		_		_			~	_	
_								_	_
							-	_	_
1	2	20, 63	-	 -			December		
	-			_			1	2	20
	number of	Message					6	7	10, 20, 63
meeten	-	-					6	11	11, 26, 63, 106, 107
MANAGE.				-			_		
2	2	10, 20, 63			******		_ 5	5	10, 11, 20, 26, 63, 106
-	eminer.		-	_			4	4	63, 107
1	1	20	2	3	20, 2 106	26, 63,	4	4	20, 26
Participal		-	_				1	1	20
1	1	26		-					
1	1	20, 108	_		_		1	1	20
	10			14				41	

1936-37 crop yielded race 63; six samples from 1937-38 crop yielded races 20 and 63

			TABLE 2 HYDERABAD				
	SAURA	SHTRA	& BARODA	· H	IYDERA	BAD	
Year	No. of stations	No. of samples	Races	No. of stations		Races	
1932-33				1	1 .	10	
1933-34				1	1,	20, 63	
1934-35	. 2	2	20, 63	_	_	-	
1935-36	1	1	10		_	_	
1936-37		-	Morte				
1937-28	_	- ,		'	_		
1928-39	2	2	20	-			
1939-40	-					_	
1940-41	3	3	20, 63	1	1	63	
1941-12	-	_	- Selection	-		_	
1912-13			-			.—	
1943-44	3	3	10, 20, 63			*****	
1914-15	4	7	10, 20, 26, 63 108	, -		. 	
1945-46	1	2	63, 107	_	-	_	
1946-47		minum j				_	
1947-48						_	
1948-49	6	6	10, 20, 26	2	2 .	10, 20, 63	
1949-50		_		_	*******	_	
1950-51	enhere.	-	_	1.	1 .	26, 106	
1951 52	_	— .	-	1	1	20	
Tota! No. of wheat samples		26			7		

Total No. of samples 1044.

-(cc	ontinu							CORE	NEPAL			
	MAD	RAS	N	AYSO:	RE	TRA	VAN	CORE		NEPA	L	
No. of sta- tions	No. of sam- ples	Races	No. of sta- tions	No. of samples	Races	No. of sta- tions		Races	No. of sta- tions	No. of sam- ples	Races	
5	6	10 :	2	2	63	_	_		2	2	10, 63	
6	7	20, 63			-	****	***	_	-	_	_	
8	9	20, 63, 107		_				_	4	. 6	20, 63, 107	
14	15	20, 63, 106, 107			_	-	-		7	7	20, 63	
19	24	10, 20, 63	-	*****				-	7	7	10, 20, 63	
16	23	20, 63	_	_	_			_	7	7	20, 63	
12	12	10, 20, 63	_	_	_	1	1	20	8	8	10, 20, 63	
15	17	20, 63			_	5	5	20, 63	-			
3	3	63	-	-	-				_	-		
1	2	106		_	_		-	-			-	
1	1	20		-		-		-		-		
4	5	10, 20, 63	2	2	20, 63	1	1	10	-	ton-en		
6	6	11, 20, 26, 63, 107	1,	3	20, 63, 108	-		No.	-		_	
		_			-		-			_		
4	4	10, 20, 63	1	1	10							
-	-		1	1	107	-	_	-	-			
2	2	11, 20, 106	-			-		-	-	-	-	
-	_		-	_	-	i			-		_	
1	1	63, 107				2	4	11, 106, 107	-	_		
7	14	11, 20, 26, 63, 106, 107	1	1	20, 108	-	_	***************************************				
	151			10			11			37		

TABLE Occurrence of physiologic races of

		KASHN	MIR	PUNJAB (including H. PRADESH)
YEAR	No. of Sta- tions	No. of Samp- les	Races	No. of No. of Sta- Samp- Races tions les
1931-32				3 3 A, 19
1932-33		Married N	party	10 10 A, 19
1933-34		-		11 12 A, 19
1934-35		Toursenth		12 13 A, 19, 31
1935-36				\$ 14 21 A, D, 19, 31 2 1 1* 19
1936-37	1	1	A, E	15 17 A, 19, 31 3 3* 19
1937-38	{2 1	2 1*	A, 19 19	(14 17 A, 19, 31
1938-39	3	4	A, 19	12 13 A, 13, 19 3 3* 19
1939-40	1	1	, A	3 3* 19 12 13 A, 13, 19 3 3* 19 8 8 A, E, F, H 1 1* 19 1 4 4 A, 19 3 3* 19 7 10 A, E, 19, 20 1 1* 19 5 6 A, F, G, 13, 20, 31 2 2* G, 19 3 3 A, F, 20 1 1* 19 2 2 13, 19 2 2 13, 19 3 6 A, D, 13, 19 1 4* 19, 20
1940-41	1	1 .	A	4 4 A, 19 3 3* 19
1941-42	-	_	_	7 10 A, E, 19, 20 1 1* 19
1942-43		-	*****	7 10 A, E, 19, 20 1 1* 19 5 6 A, F, G, 13, 20, 31 2 2* G, 19 3 3 A, F, 20 1 1* 19 2 2 13, 19 2 3* 13, 19, 31 3 6 A, D, 13, 19 1 4* 19, 20
1943-44	-	· —	-	3 3 A, F, 20 1 1* 19
1944-45				2 2 13, 19 2 3* 13, 19, 31
1945-46				3 6 A, D, 13, 19 1 4* 19, 20
1946-47	-	 , .		1 1 19
1947-48	-	-		3 7 A, E, 19, 20
1948-49	-		 -	2 8 A, 19, 20, 31
1949-50	-		-	3 8 A, H, 19, 20, 31 2 3* G, 19 3 4 A, D, 19
1950-51	-			2 3* G, 19 3 4 A, D, 19
1951-52	-			{ 15 16 A, D, E, 19, 20 4 4* 19
Total No. of wheat samples		9		189
Total No. of barley samples		. 1 .		32

3 Puccinia glumarum in different States.

DELHI	UTTAR PRADESH	BIHAR
No. of No. of Sta- Samp- Races tions les	No. of No. of Sta- Samp- Races tions les	No. of No of Sta- Samp- Races tions les
	— — — 18 18 Å, 19	 5 6 A . 19
1 1 A 1 1 31	18 18 A, 19 23 23 A, 19	, , , , , ,
		22, 22
1 1 19 ,		
1 1 10	15 16 A, E, 19, 31	5 5 A , 19, 31
1 1 19	\$\begin{array}{cccccccccccccccccccccccccccccccccccc	6 6 A , 19, 31
$\begin{cases} 1 & 1 & A \\ 1 & 1^* & 19 \end{cases}$	1 1* 19 7 8 A, 19, 31 1 1* 19	3 4 A
1 1 A 1 1* 19 1 1 A 1 1* 19 1 2 E, F	14 15 A, 19 2 2* 19	\$\begin{array}{cccccccccccccccccccccccccccccccccccc
	11 11 A, E, F, 19, 31 4 4* 19	{2 4 F, 31 1 1* 19
\$\begin{array}{cccccccccccccccccccccccccccccccccccc	15 15 Å, E, 19 3 3* 19	2 2 A, 19 2 2* 19 1 1* 19
$\begin{cases} 1 & 1 & 20 \\ 1 & 1^* & 19 \end{cases}$	7 9 A, 19, 20 3 3* A, 20	1 1* 19
$\begin{cases} \bar{1} & \bar{2} & \bar{A} \\ 1 & 1^* & 19 \end{cases}$	3 3* 19 7 9 A, 19, 20 3 3* A, 20 3 3 F, 31 1 1* G 3 4 A, 19	$\begin{cases} 1 & 1 & 19 \\ 1 & 1^* & 19 \end{cases}$
	3 4 A, 19	
1 1 19	1 1 19	
\$1 1 D	5 6 D, 13, 19, 20, 31	1 1* 19
\$1 1 D \$1 1* 19 \$1 1 19 \$1 1* G	5 5 A , 19	
1 2 31, E	3 3 A , 19	gamente discharge Migstalle
\$1 1 19 1 1* 31	\$ 4 8 A, 19, 20, 31 \$ 6 6* 31	1 1 31
$\begin{cases} 1 & 1 & 19 \\ 1 & 1^* & 31 \\ 1 & 1 & E \end{cases}$	\ \ 6 \ 11 \ \ \ \ \ \ \ \ \ \ \ \ \ \ \	1 2 A, 19
1 1 A	2 2 13, 19 4 5 A, D, E, 19	1 1* 19
	4 8 A, 19, 20, 31 6 6* 31 2 2" 13, 19 4 5 A, D, E, 19 1 1* 19 4 5 A, D, 19 7 7* 19	2 2 19
20	191	43
8	31	8

TABLE 3

						I ABLE 3
	BENG	AL AN	D ASSAM	MAD	HYA P	RADESH
YEAR	No. of Sta- tions	No. of Samp- les		No. of Sta- tions	No. of Samp- les	Races
1931-32				1	1	19
1932-33				erenin		
1933-34	_		_			_
1934-35	-	_		2	3	A, 19
1935-36			-	1	1	A ?, 31
1936-37	1	1†	31	1	1	19
1937-38	i'	1†	Α		-	_
1938-39	2	2	A, 19 .		N-PRIOR	
1939-40		-			-	
1940-41	2	2	۱A		ann and	·
1941-42				1	1	A
1942-43			_			
1943-44				1	1	19
1944-45	_		_			.—
1945-46	- Allerdonia	-				-
1946-47			 .	1	1	A, 20
1947-48		-		1	1	19
1948-49			_	_	*****	_
1949-50			_	2	2	19
1950-51	1	1	Α	-		_
1951-52			-			-
Total No. of wheat ?		7			12	
Total No. of wheat samples Total No. of barley samples						-

Total number of samples—

Wheat = 540 *Barley = 108

—(continued)

MADH (inclu	IYA BI ding I	HARAT DATIA)	RA)	JASTE	HAN	:	MAI	DRAS	NEPAL
No. of Sta- tions		Races	No. of Sta- tions	Samp-	f Races	No. o Sta- tions	Sar	np- Race	No. of No. of Sta- Samp- Races tions les
_		_	******	_	_		_		
	Toponio.		_			1	1	A	1 1 A
_			1	1	A, 19	2	2	13	<u> </u>
1	1	19	2	3	A, 31	3	3	13, 20	2 2 31
_	. —		1	1	E	. 1	1	F	7 7 A, 19, 31
-			1	1	31	5 6	6 3*	A, 13, 20 G, 19	4 4 A, 31
Marriera	-	g	1	. 3	A, 19	6 2 4 2	5 2*	A, 13, 19 19	2 2 A
-	_	-	-		_	5 2 10	6	A, 13, 19 G, 19	4 4 A, 19
_		-		_	S ERVICE STREET	7	7	A, F, 13 19, 20	,
-		_				5	5* 1	19 E	
	_	_	_	_	_	_	_		
_			-		_	٠,	_	_	
_	_	-	_	_		1	1*	19	
_			_	—		1	1	19	
	_		-	_		-	_		
		*******	-	_	_	-			
-			-		-	1	2*	G	
_	_	-	1	1	A	1	1*	G	
-		-	-		-		-		
1	1	19	{ 1 { 1	2 1*	A, 19 G	{ 1 { 1	1 1*	19 G	
1	1	19	-	-	_	2	2*	19	
	3			12			34		20
				1			27		_

TABLE 4

Occurrence of physiologic races of black and yellow rusts on grasses and Rye.

	l
	l
101	l
tritic	ŀ
	į
nin	l
rai	i
ia graminis	ŀ
III	ı
uccinia	
Ъ	
3	

Races met with	15, 40 and 42 15, and 42 40 15, 40 and 42	15 and 42 40 21 21 and 42	21, 40 and 42 40		19 19 19
No. of samples analysed	0HH00	0 H H H	, ,	ırum	
Name of the Host	Bromus patulus Unknown grass Aegilops sp. Brachypodium sylvaticum Bromus natulus	Secale cereale (Rye) Aegilops ventricosa A. triuncialis	Hilaria Jamesii Avena fatua	(ii) Puccinia glumarum	Aegilops squarossa Aegilops sp. Bromus japonicus B. catharticus Hordeum murinum Unknown grass
Name of State	Kashmir Punjab	Delhi	Uttar Pradesh		Punjab Delhi

TABLE 5

Frequency of occurrence of races of Puccinia graminis tritici

	194	1	ļ	1	-	-	1	-	1	1		}	1 :	1.0	ν.τ Σ.α	0.0	1		-	I	1
	117	-	1	1	1	1	I	1	1	1	1	1	1	2.9	χ, Ni	2.11	1.5	1.2	1	1	
	75	8.0	Σ	4.0	1.2	0.5	1	1	1	i	1]	1	1	1	1	1	1	1	1	1
Frequency of races in percentage.	42	28.3	13.3	55.7	57.3	64.0	47.9	36.8	24.5	32.9	40.0	44.08	47.8	41.9	36.3	43.9	34.8	36.2	20.0	35.5	8.5
of races in	40	23.4	33,3	15.4	16.6	0.6	18.5	46.0	53.2	40.0	10.0	21.5	15.9	14.2	1	ထ	0.8	21.2	17.3	8.0	20.2
Frequency	34	ŀ	1		1	1	t Consultation	1	1.1	တ္	17.5	1	1	1	1	1.3	1	1.2	1.3	8.0	34.0
	24	0.6	1.9	0.9	I	ľ	Expensive	1		i	2.5	1.07	1	ł	1	1	1	1	-	4.8	1.0
	. 21		6.0	1	1	*	1		1	I	1.2	1.07	1	28.6	27.3	27.8	56.8	37.7	61.3	43.5	46.9
	15	39,5	41.9	18.7	24.8	26.5	33.5	17.2	21.2	21.2	28.8	32.2	36.3	10.5	36.3	0.9	6.1	2.5		ļ	1.
Total	isolates	162	105	149	162	200	146	76	25.	85	28	94	800	105	11	. 148	132	200	77.	63	26
Total number of	samples analysed	111	78	124	124	165	102	23.5	72	64	64	63.5	. 69	20	6	837	24	. T		- TV	85
Year of	collection	1932-33	1933-34	1934-35	1935-36	1936-37	1937-38	1938-39	1939-40	1940-41	194142	1942-43	1943-44	1944-45	1945-46	1946-47	1047_48	1048-49	1010 50	1050 51	1951-52

TABLE 6
Frequency of occurrence of races of Puccinia triticina

No. of No. of samples Isolates analysed
``
22
14,
203 89
57
27
42
45
€ o
54
27
2000
41
84

TABLE 7
Frequency of occurrence of races of Puccinia glumarum

	н	1 1 1 1 1 1 1 1 1 1
	0	11.6 11.6 11.7 11.7 11.3.3 11.3.3 11.3.3
	Ĭŭ,	11.6
Frequency of races in percentage.	田	11.6 11.6 11.6 11.6 13.3 13.3 13.3
aces in pe	Ω	14.2
ency of r	4	59.6 63.6 63.6 63.6 63.6 63.6 63.6 63.0 63.0
Frequ	31	18 4444 30.6 33.7 3.7 17.0 11.1 11.1 10.4
	20	22.2 22.2 10.2 10.3 10.3 13.3 10.3 2.7
	19	88 8.0 5.0 5.0 8.0 8.0 8.0 8.0 8.0 8.0 8.0 8.0 8.0 8
	13	18.8. 18.4.2.3 18.8.6.2.3 18.8.6.2.3 18.8.6.2.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3.3 18.8.6.3 18.8.
Total	No. ot Isolates	752833274444444444444444444444444444444444
Total No. of	samples	24422222424222222222222222222222222222
Vearof	collection	1932-33 1933-34 1934-34 1934-35 1935-36 1937-38 1940-41 1941-42 1945-44 1946-47 1946-47 1946-47 1946-47 1946-47 1946-47 1946-47 1946-47 1946-47 1946-47 1946-47 1946-47

From the data presented in the foregoing tables it is obvious that some races have maintained their position of dominance from year to year and some have declined, others have shown considerable fluctuations. Regarding the races which have shown marked fluctuation, no definite conclusion can be drawn in view of the inadequacy of the samples analysed.

Shifts in population involving marked variations in the frequency of races have been recorded. Because of the altered varietal position in different regions as also change in meteorological conditions it is but expected that it would bring about change in the race flora both qualitative and quantitative. An outstanding example of the change in the rust races is that of race 56 of Puccinia graminis tritici in America. The race was picked up in 1928 but assumed first rank in 1934. On the other hand race 36 and 49 declined as race 56 increased (Stakman et al-1943). In Australia, race 34 appeared in 1926 and within a few years became a predominat race. Watson and Waterhouse (1949) have now reported that it is gradually going down. Waterhouse (1952) has recorded another such change in the population of races in Australia. According to Newton and Johnson (1946) a recent change in Canada in the racial population was the recrudescence in 1940 of races 17 which for several previous years had been of minor importance. In 1941 this race challenged the predominance of race 56 but in succeeding years it receded again into minor significance. Similar flucutations have been observed during these studies and tendency of some of the important races of the three rusts so far met with in India has been recorded below.

BLACK RUST

- Race 15: It was one of the first four races to be reported from India in collections of 1931-32 (Mehta 1933) and occupied first rank and accounted for 41.9% of the total isolates in 1933-34. It continued to be widely prevalent race till 1943-44, after which there was a gradual decline so much so that in 1949-50 it was not picked up at all. This race has been reported to consist of a number of biotypes (Loegering & Stakman-1942) but so far only one biotype 15-C has been reported in this country. In view of its high virulence there is reasonable chance that it may again assume serious proportions.
- Race 21: Which has been found in the largest number of isolates since 1947-48, was first identified in 1933-34 in a collection from Lyallpur and was not found again till 1941-42, but in 1947-48 it was found in 56.8% isolates and in 1949-50, in 61.3% and in 1951-52 in 46.9%.
- Race 40: Was one of the first races to be isolated in India. It attained first position in prevalence in years 1938-39, 1939-40 and 1940-4! and has been found to occur every year throughout this period of twenty years, like race 42.
- Race 42: It was first isolated in combination with races 40 and 75 from 1930-31 crop and has been found every year since then attaining the first rank during the periods 1934 to 1938 and again

1941 to 1947. This is the only one amongst important Indian races to which the *dicoccum* wheat Khapli is susceptible.

Race 75: This race requires particular mention because of its complete absence since 1936-37. It was one of the four races originally picked up in two collections of 1930-31 in combination with races 40 and 42. Although it is difficult to explain the absence of race 75, it is interesting to point out that the reactions of differential hosts to this race are covered up by those of races 21 and 34. Since race 21 has become the most prevalent race it is quite possible that it masks the reactions of race 75 so completely that it becomes extremely difficult to detect the presence of the latter in a mixture.

The fluctuation in the occurrence of the four most common races is shown in the fig. (p. 249).

Brown Rust: Mehta (1933) reported the occurrence of races 10 and 63. Race 20 was first picked up from 1932-33 collections. These 3 races have continued to occur almost every year. Race 63 has been found to occur in the largest number of collections followed by race 20. The remaining five races viz. 11, 26, 106, 107 and 108 have been met with only occassionally in different years. In 1947-48, however, race 26 accounted for 25.9% of the isolates and stood second. Mediterranean and Democrat, two of the differential hosts, have been found to be resistant to all the Indian races of brown rust and have so far provided valuable material to the plant breeder for evolving rust resistant varieties. They are, however, susceptible to the recently discovered race 77.

Yellow Rust: Race 19 and a new race "A" were the first to be identified in order of prevalence. Out of the remaining races, 13, 20 and 31 have been picked up over a large number of years. Race 31 was, however, comparatively more prevalent during 1934-35, 1935-36 and 1936-37 but races D, E, F, G and H have so far been rare.

MAINTENANCE OF RACES

Single spore cultures of all the races described above are being maintained at the Rust Research Sub-Station, Simla (App. 6,900 ft. a.s.l.). The primary aim of maintenance of cultures is to ensure the availability of inoculum of any race for testing the varieties in the glasshouse as well as in the field. The cultures are maintained on a susceptible variety in double layered muslin chambers. To avoid chances of contamination no two races of the same rust are repeated on the same day. No difficulty has been experienced in maintaining the cultures of black and brown rusts throughout the year but for yellow rust cooling of the glasshouse has at times been found necessary in summer months,

Some of the cultures have completed more than 300 generations without any apparent loss in virulence. So far no case of mutation involving change in pathogenicity has been observed. Colour mutation has however, been observed recently in races 15-C and 194. The history of all the races maintained at Simla sub-station is provided in table 8.

TABLE 8

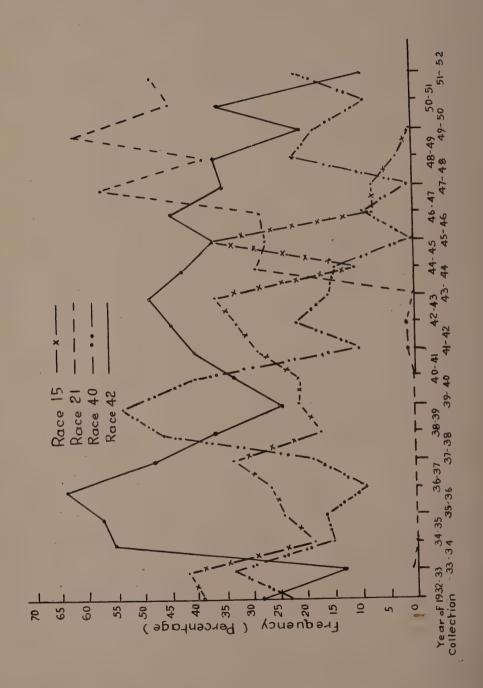
History of Single Spore Cultures of the three rusts upto June, 1953.

(i) Puccinia graminis tritici

Age in generations	274 250 262 182 309 308 317 102		314 109 270 107 304 259 259
Started in	March, 1934 June, 1935 April, 1935 October, 1940 May, 1932 July, 1932 December, 1931 October, 1945 October, 1945	a	December, 1931 August, 1945 May, 1935 August, 1945 December, 1931 July, 1935 September, 1935 October, 1935
Stock collection or isolation	Stock Ver. (2)-Ko.(4) Ver. (2)-Khp. (2) Ko. (4) Enk. (1) Khp. (4) Stock Ver. (4) Acm. (1)	(ii) Puccinia triticina	Stock Lor. (3) Hu. (4) Hu. (4) Stock Lor. (3)-Br. (4) Hu. (3) Web. (2)
Name of Station and Original Host	Ketty—Local Lyallpur—C.518 Himayatsagar—Bansi Pusa—14-10-1 Poona—Pusa-4 Poona—Pusa-4 Simla—Local Tharsa—No.281 Betul—Mixed Vars.		Lyallpur-Local Anikorai—Local Choharpur-Local Lingmala-Var.? Simla-Local Haldwani-Local Khanewal-Pb. 8A. Banaras-Pusa-4
Race	15 22 46 42 117 117 194 194		10 11 20 26 63 63 106 107

(iii) Puccinia glumarum

283 284 284 320 284 284 247 182	
November, 1935 October, 1933 December, 1935 November, 1935 January, 1934 April, 1936 February, 1937 January, 1941 January, 1941 January, 1943	
Stock Stock Web. (4) Ch. (4) Web. (3) Spal. (2-3) Spal. (2-3) Spal. (2-3) Pet. (3) Hein. (3)	
Thambatti—Local Fyzabad—Local Thambatti—Local Dehradun—Local Narkanda—Local Kangra—Pb. 17 Barabanki—P. 4 Ketty—Local Anikorai—Barley Local Rawalbindi—Local	
EUSSEADH FRE	



DISCUSSION

From examination of the results for the last twenty years certain conclusions can be drawn. Most of the races e.g. 15, 21, 40 and 42 of black rust, 10, 20 and 63 of brown rust and races 19 and 'A' of yellow rust which were first to be identified in this country have been prevalent almost every year. The remaining races which were detected later do not so far appear to have assumed such importance from the point of view of distribution. For obtaining more accurate information it would be necessary to analyse samples obtained from all the wheat growing tracts in the country, the number of samples depending on the intensity of cultivation keeping in view the varieties involved. The gradual disappearance of race 75 and the sudden rise of race 21 however, seems difficult to explain. There is little information about the factors which are responsible for the changes in the physiologic race flora of a country. It has been stated by Loggering (1951) that environmental factors exercise a marked influence on the survival of races and that this effect varies from race to race.

The development of different races would probably depend on the fluctuations or range of temperature available during the rust development phase as also combination of different environmental factors during the pre- and post infection periods in relation to the varieties involved. Intensive critical study involving these factors individually is no doubt difficult and long drawn but is essential to obtain a true picture.

Earlier, the occurrence of six races of black rust, six of brown rust and eight of yellow rust had been reported. Since then four more races of black and two each of yellow and brown rusts have been recorded. The origin or the appearance of new races is difficult to explain because of the fact that the functional aecidia of black and brown rusts of wheat have not been observed so far. These races might, therefore, have come into existance either as a result of the alternate hosts functioning in some undiscovered locality or by mutation. It is probable that these races might have existed before and their presence was not detected due to poor distribution and also due to the fact that comparatively fewer samples were analysed in the past. Although the dissemination of rusts from other countries to a geographically secluded sub-continent like India, by means of wind-currents, does not appear to be normally feasible, the possibility of chance introduction of spores and of a new race thereby cannot be ruled out altogether.

Ten races and three biotypes i.e. 15, 21, 24, 34, 40, 42, 75, 117, 122 and 194, and 15-C, 42-A, and 42-B of Puccinia graminis tritici have been found. Races 40 and 42 were met with throughout the period of this study, races 15 and 75 have gradually declined. On the other hand race 21 which was found only once in 1933-34 and then again in 1941-42 has become most predominent in recent years. Other races have shown marked fluctuations in frequency from year to year.

Out of eight races of *P. triticina* races 10,20 and 63 have been picked up almost every year. The remaining five races viz.

11, 26, 106, 107 and 108 have been met with only occassionally in different years.

Amongst the races of *P. glumarum* races 19 and 'A' have been found every year and in the largest proportion of isolates, races 13, 20 and 31 have been picked up over a large number of years whereas the remaining races D, E, F, G and H were extremely rare. Race G appears to be chiefly restricted to barley.

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A NEW COLLETOTRICHUM FROM INDIA

M. V. DESAI AND N. PRASAD.

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During the rainy seasons of 1952 and 1953, a peculiar disease was observed on guar (Cyamopsis tetragonoloba Taub.) at the Agricultural Institute, Anand. There are two types of guar cultivated in this region, one is known as the hairy variety and the other glabrous. The former is used as fodder, while the latter is used as vegetable. This disease was mostly found on the glabrous types and was characterised by the appearance of black spots on the stem, petioles, and leaves. Even in advanced case of attack, no necrosis of any part of the plant occurred (Fig. 1).



Diseased

Fig. 1

Healthy

Symptoms of the blight of guar (Glabrous type) in nature

Cross sections of the affected parts of the host revealed the presence of fungus hyphae and at certain places acervuli with brown setae and spores were found. The shape of spores varied from cylindrical to elongated oval or sickle shaped.

MATERIALS AND METHODS

Isolations were made from the affected tissues in the usual manner and almost in every case, a fungus with submerged black to olive brown mycelium with acervuli and setae was obtained. The culture was single-spored following the methods of Hansen and Smith.

Infection experiments were conducted by using the pure culture. Infection was carried out in two ways. (i) The seed was dipped in spore suspension and then sown in 9" pots filled with sterilized soil, (ii) a spore suspension of the organism was atomised on three week old seedlings of guar. In the first method, black streaks on the stem were observed ten days after the emergence of the seedlings. In the second method typical symptoms were observed six or seven days after inoculation vide Fig. 2.

A number of hosts were inoculated for finding out the host range of the parasite. Among the hosts tried, were Crotalaria juncea L., Sesamum orientale L., Cassia tora L., Crotalaria retusa L., Cajanus cajan L., Capsicum annum L., and Gossypium herbaceum L. Each one of the above mentioned hosts was inoculated in two ways described above. This organism failed to infect any host other than Cyamopsis tetragonoloba.

THE ORGANISM

The fungus grows well on potato dextrose, Richard's and oat meal agar media.

It is characterised by the presence of mycelium which is dark olive green to black in colour. The hyphae are septate, containing oil globules and are 4.0 μ to 6.6 μ in diameter. Abundant acervuli are formed, with a size varying from 77 μ to 205 μ in diameter. The setae are irregularly arranged, dark purplish throughout and are $54.3\times7~\mu~63\times8.8~\mu$. The conidia are borne singly, hyaline, but pale pink in mass like sporodochia, non-septate, shape varies from cylindrical to elongated oval or sickle shaped, chlamydospores both intercalary and terminal. A type culture of the fungus is deposited with the culture collection at the Indian Agricultural Research Institute, New Delhi.

The organism possesses a thick stroma, setae and falcate spores (vide Fig. 3). These characters place the fungus under the genus Colletotrichum. The form genera Colletotrichum and Glaeosporium belonging to the group Melanconiales are very difficult to separate into species and morphologically very variable and are generally weak parasites, a single species infecting and producing diseases in a great variety of host plants as shown by Shear and Wood. Ling and Lin (1944) state that in comparison with a number of species of

Colletotrichum such as C. circinnans (Berk.) Vogl., C. indicum Dast, C. truncatum (Schw. Andrus and Moore) and Glomerella glycinis (Hemmi) Lehman et. Wolf, C. capsici (Syd) Butl. et. Bisby, differs from them in no essential way.



 $\label{eq:Fig.2} \textbf{Fig. 2}$ Symptoms of the disease under artificial inoculation

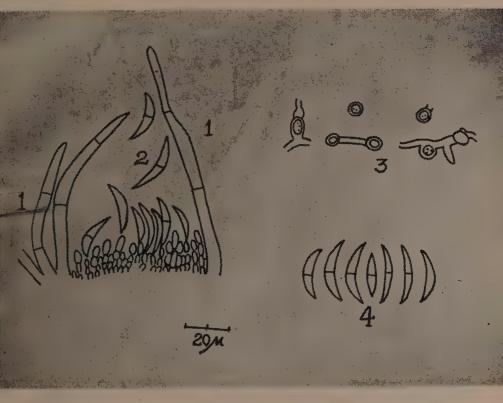


FIG. 3

Colletotrichum capsici f. cyamopsicola n. f. 1. setae. 2. & 4. spores. 3. chlamydospores.

Butler (1918) has shown the extreme variability in the dimensions of acervulus within the species $C.\ capsici$ as ranging from 75-120 μ . The acervulus of the organism under study varies from 77-205 μ . Ramakrishnan (1947) found similar variation in the size of setae as well. Our study too has shown a great range of variability in this character.

The only dependable character for taxonomic considerations appears to be the shape and size of conidia. The conidia in the genus Colletotrichum are known to be either oblong, spindle shaped or falcate with tapering or blunt ends. Their size has been known to be influenced by the substrate but it varies within limits. With the result, this character has been utilized for differentiation of species, Judging by all considerations, the organism under study does not seem to agree with any species of Colletotrichum described so far. Shear and Wood (1907), Ling and Lin (1944) and Dastur (1934) have laid great emphasis on pathogenicity on different hosts in differentiating

species within this genus. *C. capsici* was first recorded on *Capsicum* to which host it owes its specific name. Dastur (1934) created a provisional new species of *C. indicum* causing seedling blight of cotton. The only difference between his isolate and *C. capsici* was that of pathogenicity. *C. indicum* did not infect capsicum nor *C. capsici* did infect cotton. Recently Patel, Kamat and Pande (1952) have created new species *C. crossandrae* on the basis of its pathogenicity.

Recently Tiffany and Gilman (1954) have made a study of the genus Colletotrichum and have divided the genus into two groups viz. (i). curved spore group and (ii) straight spore group. According to them our isolate would fall under Colletotrichum truncatum (Schw.) Andrus and Moore. This species has been recorded on stem and pods of Phaseolus lunatus L., P. vulgaris L., Medicago sativa L., Melilotus alba Desr., Trifolium pratense L., Glycine max (L.) Mew, Vicia villosa Roth and Lotus purshianus Clem and Clem. The size and shape of acervuli, setae and conidia seem to fall within the limitations described for this species but our isolate has failed to infect any host other than Cyamopsis. Even among the two types of guar reported from this area, it shows a definite preference for the glabrous variety. On different species of the genus Phaseolus grown in India, we find that Colletotrichum lindemuthianum occurs very commonly. It can be easily distinguished from our isolate as it belongs to the straight spore group.

Wiltshire in a personal communication suggested that Colletotrichum isolated from cluster beans belonged to Colletotrichum capsici (Syd.) Butl. and Bisby. Cross inoculation tests carried out on other hosts mentioned above conclusively proved that the isolate was specialised in its parasitism on the genus Cyamopsis only. Doidge (1952) has recorded a Colletotrichum on the genus Cyamopsis from South Africa but has made no determination of species. Wiltshire in his communication mentioned that he has not seen anything like it on the genus Cyamopsis. Considering the very specialized parasitism obtained in our isolate, it appears the best way to deal with the situation would be to recognise it as a new form under the species C. capsici.

Colletotrichum capsici f. cyamopsicola forma Nov.

Mycelium submersum, Fusce vel olivacee viride, haud copiosum; hyphae septatae, continentes globulos olerosos, magnit, $4.0-5.6~\mu$ diam., acervuli abundantes, magnitud. Variabilis $77-205~\mu$ diam. setae irregulariter dispositae, fuace purpurascentes, $54-63\times7-8.8~\mu$; conidia singula, hyalina, pallide roses in massa similia sporodochus, ut plurimum uniseptate, formae variabilities, cylindrica vel elongato-ovate vel falcate, conidia 0-septate $17.5-28.0\times3.5-5.2~\mu$. Inficit Cyamopsidem tetragonolobum Taub.

Typus lectus in urbe Anand, mense Auguste anni 1952 at positus in Plant Pathological Herbarium, Agricultural Institute, Anand, positus etiam Indian Agricultural Research Institute, New Delhi, atque in Commonwealth Mycological Institute, Kew in Anglia.

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DECLINE IN CASHEWNUT

T. S. RAMAKRISHNAN

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Introduction

Cashewnut (Anacardium Occidentale L.) is cultivated extensively on the coastal regions of South India both on the western and eastern sides of the peninsula covering about 40,000 acres in extent. The enhanced prices now prevalent for the kernel have given a fillip to the further extension of its cultivation. However, the plantations are raised invariably in dry waste lands of low fertility where other annual crops do not come up well. The lateritic undulating lands and hillocks of the west coast and the sandy stretches of the seacoast on the east and west coasts are planted with this tree. Nor does the tree receive much attention either in cultural operations or in manuring (Patel, 1932). Close planting is practised and the trees form a source of fuel also. Till recently the nuts formed a secondary source of revenue but with the high prices now prevailing for the kernel they have gained in importance and have become of primary value.

Despite the poor attention, the plantations continue to yield, though erratically, for years. As casualties occur among the trees they are cut down to be sold as fuel. The greater interest now evinced in this crop has revealed the existence of several diseases affecting the yield and duration of the trees. Plant protection measures are beginning to be adopted in some of the commercial plantations to combat these diseases while in most of the others no attempt is made to check them.

COMMON DISEASES

Only a few diseases have been recorded on this host previously from South India. The most important of these is the 'pink disease' caused by *Pellicularia salmonicolor*. This is prevalent on the west coast and brings about the 'die-back' of branches (Sundararaman 1932). Patel, Kulkarni and Moniz (1948) have obtained successful infection of cashew leaves with *Pseudomonas mangiferae-indicae* isolated from mangoes.

During the last decade, heavy incidence of powdery mildew has been prevalent in the neighbourhood of Calicut and in Cochin state resulting in the shedding of flowers and drying up of the inflorescences. Viegas (1945) has reported the occurrence of Oidium anacarati on cashew from Brazil. A grey blight associated with Pestalotia virgatula Kleb, has been observed in parts of Malabar and South Kanara. But it appears that the organism is only of secondary importance and affects only leaves weakened by other causes like sun scorch or lack of nutrition. A twig blight is reported from

plantations near Trichur and Calicut causing the drying of the terminal portions of the branches. *Gloeosporium* sp. is associated with this disease. Here again it is interred that the fungus is only of secondary significance, other soil and nutritional defects being the chief factors. Lesions on the leaves caused by *Gloeosporium* sp. have been observed from Amazonia (Deslandes 1944).

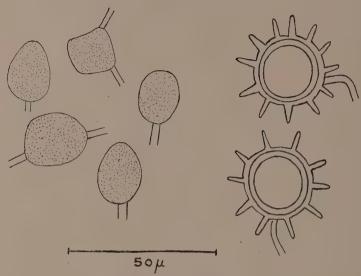
SYMPTOMS OF DECLINE

During the last few years a type of decline of cashew has been noticed in some of the plantations near Calicut. Some of the branches become defoliated during the summer months and the twigs dry up. Gradually this phenomenon is exhibited by more and more branches and in the course of two or three years the tree dies. Young and old trees are affected in this manner. The symptoms indicated that there was something wrong with the root system and that the absorption of water was insufficient. The decline is prevalent in the plantations on the lateritic undulating lands and hillocks in the neighbourhood of Calicut.

Examination of the roots of the effected plants showed that many of the finer fibrous roots had rotten and were dead and dark brown in colour. Non-septate hyphae and oospores of *Pythium* were observed in the tissues of such roots. It was suspected that the decline may be due to the infection of the finer roots by the pathogen. This is comparable to the decline of avocadoes caused by the infection of the feeder roots by *Phytophthora cinnamomi* Rands (Zentmyer and Klotz 1947). The symptoms of decline in cashew were accentuated from the time of the cessation of monsoon rains and became severe in the dry summer months.

THE PATHOGEN

Specimens of the affected roots were collected from selected trees exhibiting symptoms of decline and isolations of the organisms were made. Invariably growths of a Pythium were obtained. The cultures were purified and the characters of the fungus were studied. The fungus grew luxuriantly on oat agar. A white cottony arachnoid growth was obtained which filled up the dishes in three to four days. The hyphae were coenocytic, branched and hyaline, measuring 3 to 5μ in diameter. Asexual reproduction took place by the formation of sporangia, developed acrogenously or intercalarily. These were spherical, hemispherical or spindle-shaped and very variable in size. They germinated like conidia producing germ tubes. Sexual reproduction was also abundant. The oogonia were spherical, terminal or inter-calary and with several conical obtuse spines on them. The average diameter of the oogonium excluding the spines was 18µ (14 to 28). Normally one monoclinous antheridium was attached to each oogonium. The oospore was spherical, smooth and plerotic with an average diameter of 16μ (12 to 26). Mature oospores were of a light yellow colour. Germination of the oospore was obtained after 24 to 48 hours when mature ones were floated in water. A germ tube grew out piercing through the outer persistent oogonial wa 11.



Sporangia and oospores

PATHOGENICITY

The parasitism of this isolate was tested on cashew by inculating the soil round the collar region of healthy six months' old seedlings growing in pots. There was no evidence of any infection even after two months.

Since the pathogen was observed to be infecting in nature only the finer roots the method of inoculation was modified in such a manner as to place the fungus culture directly in contact with the finer roots. Healthy plants growing in tile pots were selected. The disc blocking the bottom of the pot was removed and the soil sprayed with water to expose the finer roots near the bottom. The culture of the pathogen was placed on the exposed roots and the soil and the disc replaced in position. Control plants were similarly treated but the culture of the fungus was not added. In the course of three weeks the inoculated seedlings began to exhibit symptoms of wilting. A week later all the inoculated plants died preceded by detoliation. The fibrous roots of the affected plants were rotten and the same fungus was reisolated from them. The control plants remained healthy and had put forth fresh leaves. This indicates that this Pythium is capable of causing root rot of cashew and consequently may be one of the factors responsible for the decline. Since the fungus thrives in moist soil the infection must be taking place during the rainy season but the effect becomes apparent during the dry weather.

IDENTITY OF THE FUNGUS

The presence of spines on the oogonium and the plerotic nature of the oospore indicate the affinities of the isolate to a small group of species consisting of P. acanthicum Dresch, P. mamillatum Meurs., and P. spinosum Saw. (Middleton 1945). The shape of the sporangia, the antheridia, the spines on the oogonia and the measurements of the oogonia and the oospores indicate that the isolate has to be identified as P. spinosum.

TREATMENT

Two trees which had exhibited initial symptoms of decline were given the following treatment. The soil at the base of the tree was forked and drenched with Cheshunt compound solution (one ounce in two gallons of water). Later each tree was manured with 100 pounds of compost, 2 pounds of ammonium sulphate and $1\frac{1}{2}$ pounds of superphosphate incorporating them with the soil in June. Six months later the trees had revived with plenty of green foliage while other trees which had exhibited similar symptoms of decline in June had deteriorated. Though the treatment is of an empirical nature the results indicate that the decline may be averted by proper and adequate soil amendments. Zentmyer and Klotz (1947) state that treatment of the soil from decline-affected avocado plantations in America with chloropicrin, ethylene dibromide or steam rendered it free from *P. cinnamomi*. But these cannot be used in existing plantations.

Although *P. spinosum* was pathogenic on cashew roots, field observations indicate that it may not be the sole cause of the decline. It has been stated earlier that cashew plantations are raised in poor infertile lands. The nutrient status of the soil may not be sufficient to maintain the trees in robust conditions for many years, though it must be admitted that cashew is one of the hardiest of the plantation crops and can stand a certain amount of neglect. Nevertheless the addition of organic matter and other fertilisers will improve the condition of the plants and enable them to resist infection. The organic matter will further improve the soil condition and encourage the multiplication of saprophytic organisms (e.g., Trichoderma viride Pers.-T. lignorum (Tode) Harz) which will be helpful in keeping down the pathogen. T. viride has been found to parasitise many species of Pythium.

However it may be questioned whether it will be economical to carry out these manurial and cultural operations to cashew trees. Hitherto the expenditure incurred on the maintenance of these plantations has been practically negligible. Most of the growers are averse to adopt these measures. But one cannot expect the trees to grow and yield for a long time if adequate nutritional requirements are not provided. Improvements in the cultural operations are necessary especially for the plantations on hill slopes to prevent soil erosion and leaching away of nutrients from the soil. The grower has to choose between adequate manuring and other operations with the

resultant improvement in the growth of the trees and the yield of nuts or continuing the present practice and accept the low yield and the early decline of the trees.

SUMMARY

In addition to the enumeration of the diseases affecting cashew in South India, the symptoms of a decline of the trees in the west coast are described. The affected trees exhibited rotting of the finer roots. *P. spinosum* was isolated from the diseased roots and this was found to be pathogenic to the roots of cashew and causing the death of young plants. Drenching the soil with Cheshunt compound and application of manures arrested the progress of decline in the treated trees.

I am thankful to Srimathi C. K. Soumini for help in the isolation of the fungus.

R. S. Puram, Coimbatore.

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EXPLANATION OF PLATE

- Fig. 1. Inoculated cashew plants (two) on the left and the control on the right.
- Fig. 2. Germinating oospore of P. spinosum.

PLATE I



FIG. 1



FIG. 2

STUDIES ON THE ANTAGONISTIC ACTINOMYCETES FROM THE SOILS OF WEST BENGAL,

S. K. MUKHERJEE AND P. N. NANDI

(Accepted for publication, August 28, 1955)

INTRODUCTION

Although the actinomycetes form a large and important group of micro-organisms in soil, very little attention was paid to them before the discovery of streptomycin. Subsequent discovery of chloromycetin, aureomycin, terramycin and other antibiotics elaborated by the above group of micro-organisms greatly stimulated the search for the newer types of antagonistic strains of actinomycetes.

Extensive survey of the antibiotic-producing soil microflora was, therefore, undertaken by workers all over the world and it has been held that antagonistic actinomycetes are widely distributed in nature (Benedict, 1953).

In India, information is lacking on the systemtic survey of antagonistic actinomycetes. Mukherjee et al (1954) isolated 235 strains of actinomycetes from the soils of various parts of West Bengal and tested 121 strains against both gram-positive and gram-negative test bacteria. Bhide et al (1952) tested 64 strains of actinomycetes against 20 spp. of Xanthomonas (plant pathogen) of which 7 proved to be inhibitory to 3 or more test bacteria while nearly all the test organims were inhibited by at least one strain of antinomycetes. Chakraborty et al (1952) isolated 40 strains from the soils collected in the neighbourhood of Calcutta and reported that 8 strains of actinomycetes inhibited all the 4 test organisms (Staphylococcus aureus, Escherichia coli, Eberthella typhosa and Vibrio cholerae) used, while 17 strains of actinomycetes were found to be antagonistic to at least one test organism. In the present investigation, a detailed survey of the antagonistic actinomycetes from the soils of West Bengal is reported.

MATERIAL AND METHODS

Soil samples were collected from different parts of West Bengal from cultivated fields, gardens, fallow lands, composts and from virgin plots. The samples were taken from a depth 3" below the surface level. Soil samples containing excessive moisture were air-dried, pulverised, sieved through a 3 mm. mesh and stored in dry tin cases. Dilution plates were prepared within one week from the date of collection. 10 gms. of sieved soil were suspended in 90 ml. sterile tap water in 250 ml. Erlenmeyer flasks, shaken thoroughly and allowed to stand for 20-30 minutes. Dilutions in steps of 10, up to a strength of 1:10,00,000 were made in 9 ml. sterile tap water blank aud suspensions were plated out in triplicate in Thornton's and Norris' media

(sol. starch—2.0 gm., asparagine—0.05 gm., MgSO $_4$ -0.20 gm., CaCl $_2$ -0.05 gm., FeCl $_3$ -0.01 gm, NaNO $_3$ -0.05 gm., Distilled water to 1000 ml. Agar agar-2%, pH—6.8). The plates were incubated at 24-25°C for 15 days and then isolations were made.

The actinomycetes thus isolated from soil were put to pure culture and continued in sub-culture in potato dextrose meat extract agar slants (Kelner and Morton, 1947) for future use. The following test bacteria and fungi were used in the assay work—(1) Staphylococcus aureus. (2) Escherichia coli. (3) Eberthella typhosa. (4) Vibrio cholerae. (5) Alternaria solani. (6) Curvularia specifera. (7) Helminthosporium oryzae. (8) Fusarium sp. and (9) Rhizoctonia sp. Primary screening of actinomycetes was carried out by agar-streak method (Waksman and Reilly, 1945). Preliminary investigation was carried out with regard to the efficiency of the production of antibiotics in the following media:

- (1) Beef ext. sodium chloride medium (Waksman and Schatz, 1946).
- (2) Soybean meal medium (Rake and Donovick, 1946).
- (3) Potato dextrose meat extract medium (Kelner and Morton, 1947).
- (4) Straw infusion medium (Dey, 1947).

Of these, straw infusion medium was generally found to give better results than the rest in stationary culture. In the present investigation, modified straw infusion medium was used and the composition of the medium is given below.

Modified straw infusion medium

Tryptone (Difco)	 2.50 gm.	K_2HPO_4	2.04 gm.
Lab, lemco	 1.00 gm.	FeSO ₄ .	0.01 gm.
Glucose	 20.00 gm.		0.01 gm.
NaCl	 0.50 gm.	10% straw infusion .	500 ml.
MgSO ₄ 7H ₂ O ₄	 0.25 gm.	Tap water to 1000 m	i.

pH-7.2

Straw infusion was prepared by keeping overnight 100 gm. of paddy straw in 1000 ml. of water at room tempreture (26°-28°C). The infusion was filtered through filter aid on a Buchner funnel under reduced pressure. Tryptone was found to be a better substitute for peptone and addition of beef extract was useful. To test the antagonistic properties of actinomycetes, equal volumes of straw infusion agar and nutrient agar were mixed thoroughly, poured into plates and the usual agar streak method of assay was followed. The plates were incubated at 37°C and the degree of inhibition, if any, was recorded up to the nearest millimeter. In the case of pathogenic fungi, the method adopted was essentially the same as in the case of bacteria. Czapek-Dox's agar, adjusted to pH 7.2 was used instead of straw infusion agar. The streaks were drawn with conidia of sporesuspension of the test fungi in sterile water prepared by transferring a loopful of spores from 4-day agar culture.

The actinomycetes possessing marked antagonistic properties were further assayed by agar-cup method (Waksman, 1947). They were grown in 250 ml. Erlenmeyer flasks containing 50 ml. of straw infusion medium for 8 days, at 28°C. The bacterial test organisms were grown on nutrient agar slants, loopfulls of fresh cultures (grown overnight) were transferred to nutrient broth and the suspension was used after incubating for 2 hours at 37°C. 25 ml. quantities of nutrient agar were distributed in sterile petri-dishes (dia. 9 cm.) which were seeded with a hundreth dilute suspension of test organisms. Plugs were punched off with a sterile cork-borer (dia. 8 mm.) 0.1 ml. of the culture filtrate was poured into each cup carefully and the plates were incubated overnight at 37°C. Diameter of the zone of inhibition was measured up to the nearest millimeter. When the pH of the culture filtrate was found below 6, it was adjusted to neutrality by adding sterile 5% NaHCO3 solution. The same procedure was adopted in the case of fungi and potato dextrose agar was used instead of nutrient agar.

RESULTS

Of 242 strains of actinomycetes, isolated from 63 soil samples, 170 strains were tested for antagonistic properties. Of them, 68 strains (40%) were inhibitory to gram-positive bacteria, 38 strains (22.3%) to gram-negative bacteria and 54 strains (31.7%) to fungi. Among the cultures tested, 26 strains (15.2%) were strongly inhibitory to gram-positive bacteria, 19 strains (11.1%) to gram-negative bacteria and 7 strains (4.2%) to fungi (Fig. 1).

Among the active strains, 50 were further tested both by agarstreak (Fig. 2) and agar-cup (Fig. 3) methods using the same test organisms described above. The actinomycetes strains were identified according to the methods given in Bergey's Manual of Determinative Bacteriology (1948) as also by the method of Pridham and Gottlieb (1948) based on the utilisation of different carbon compounds. The results of antibacterial and antifungal tests are given in Table 1.

DISCUSSION

From our results it is noticeable that agar-streak method seems to be more sensitive in general than the agar-cup method of assay (Routein and Finlay 1951). Among the gram-negative bacterial test organisms E. coli was very susceptible while two others e.g., Eb. typhosa and V. cholerae showed resistance to the action of antibiotics produced by the actinomycetes. Of the plant pathogenic fungi, Helminthosporium oryzae and Curvularia specifera were found to be greatly inhibited while Fusarium sp. remained insensitive to a certain extent to the action of antibiotics. It appears from Table 1 that S. erythrochromogenes is particularly active both against gram-positive and gram-negative bacteria. Erythromycin has recently been isolated from culture filtrate of a strain related to this species (Mc Guire et al 1952). No antibiotics have, however, so far been isolated from S. flavovirens. S. erythreus. S. rutgersensis and S. griseolus—some of which have been shown to possess remarkable antibiotic properties

Antibacterial and antifungal spectrum of different Streptomyces spp. isolated from the soils of West Bengal (Inhibition zone in millimeter) TABLE 1

Test Organisms.

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S.M. = Agar-Streak Method; C.M. = Agar-Cup Method; + = Positive inhibition; - = Negative inhibition. * = Indicates related species.

TABLE 1 (Continued)
Test Organisms.

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S. rulgersensis 28. AC ₂ (177) 29. AC ₃ (183) 30. AC ₄ (195) 31. AC ₂ (197)	S. albidoflavus 32. AC _s (177)	S. antibioticus *33. AC ₂ (185)	S. griseolus 34. AC ₃ (194)	S. scabies *35. AC ₁₁ (195)	S. lavendulae *36. AC _s (196)	S. purpeochromogenus 37. AC ₁₂ (217)	Streptomyces spp. (unidentified) 38. AC ₁ (9) 39. AC ₁ (60) 41. AC ₁₈ (BG.) 42. AC ₁₈ (177) 42. AC ₁₈ (183) 44. AC ₁ (193) 44. AC ₂ (193) 45. AC ₂ (193) 46. AC ₃ (203) 47. AC ₄ (203) 48. AC ₄ (203) 50. AC ₆ (217)

S.M.=Agar-Streak Method , C.M.=Agar-Cup Method ; +=Positive inhibition ; -=Negative inhibition. *=Indicates related species.

in the present investigation. Some of the unidentified Streptomyces which differ from all known forms and exhibiting remarkable antagonistic action against all test organisms merit detailed investigation.

Though most of the antibiotic substances produced by Streptomyces strains investigated possessed antibacterial properties, fewer of them inhibited plant pathogens. The strains AcI2 (217) of S. purpeochromogenus and certain spp. related to S. griseus and S. albus deserve special mention in this connection. The isolate ACI (214) identified as S. viridochromogenes is active only towards fungi.

SUMMARY

- 1. 170 strains of actinomycetes were isolated from the soils of West Bengal and tested against pathogenic bacteria and fungi. Of them, 40% were found to be inhibitory to gram-positive bacteria, 21.1% in gram-negative bacteria and 31.7% to higher fungi.
- 2. Among the test organisms, Staphylococcus aureus and Helminthosporium oryzae were fairly susceptible to antagonistic actinomycetes, while Vibrio cholerae and Fusarium sp. were relatively resistant.
- 3. Different isolats belonging to the same species possessed different antagonistic properties.
- 4. Some strains which exhibited antagonistic action when tested by agar-streak method, failed to do so by agar-cup method.

Thanks are due to Dr. D. M. Bose, Director, Bose Research Institute for help and encouragement in the work.

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PHYTOPATHOLOGICAL NOTES

Occurrence of Ustilaginoidea Virens (CKE.) TAK. on Oryza Officinalis Wall—P. Govinda Rao and T. C. Venkata Reddy. Ustilaginoidea virens (Cke.) Tak. causes the false smut or the green smut disease in paddy in most of the rice growing parts of the world. The disease has also been reported from India by Butler (1913), Raychaudhuri (1946) and Padwick (1950). It appeared as an epiphytotic in the first crop paddy season in November, 1953 in the coastal districts of Andhra, causing considerable economic loss.

During January, 1954, a wild rice, Oryza officinalis Wall grown in the Agricultural Research Station, Maruter in the West Godavari district was found to have been infected by false smut. A few grains, about ten, in each panicle were infected. The glumes remained uninfected and the ovaries were transformed into roundish to elliptical green masses. The sclerotia are longer along the axis of the grain and measure 3.0 to 5.3×1.0 to 2.0 mm. average (of 70 selerotia) being 4.1×1.4 mm. A cross section of the sclerotium showed yellowish central portion surrounded by a blackish green layer which is powdery and consisted of mature spores of the fungus. Spores are round, olive brown in color, verrucose and measure 3.9 to $7.1~\mu$ in diameter, average (of 200 spores) being $4.9~\mu$. The younger spores are small, pale in colour and almost smooth. Spores did not germinate in water or in 1% sucrose solution. The fungus could not be brought into culture.

Macroscopic and microscopic characters of this fungus and that of U. virens on oryza sativa are given below.

Oryza Sativa

Sclerotia: Whitish in centre followed by orange yellow and dark green towards outside.

 7.8×48 mm. $(5.5-10 \times 3-8$ mm.)

Spores: Mature spores olivaceous and warty, young spores smooth and pale.

 $4.2-6.3 \mu$ in diameter average 4.9μ

Oryza officinalis

Yellowish in centre surrounded by dark green layer.

 4.1×1.4 mm. $(3-5.3 \times 1-2$ mm.)

Mature spores Olive brown and verrucose young spores almost smooth and pale.

 $3.9-7.1 \mu$ in diameter average 4.9μ

From the above data it is observed that the spores produced by these fungi are of nearly the same size. The sclerotia of wild rice are smaller than those produced on the cultivated one and the internal white colour produced in the sclerotia of the latter is absent in the former. The size of the grain in O. officinalis is smaller, than

in O. sativa and that might be the reason for the smaller sclerotia. The internal Colour produced in the sclerotia may vary with the species or variety of the paddy. Hence the fungus causing the false smut in O. officinalis is also considered to be Ustilaginoidea virens (Cke.) Tak.—Agricultural College, Bapatla (Andhra State).

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A new physiologic race of Puccinia triticina Eriks, in India—R. S. Vasudeva, V. C. Lele and D. P. Misra. During analysis of wheat rust samples from the crop of 1953-54 a new physiologic race of leaf rust of wheat hitherto unrecorded from India has been isolated from the samples received from Bihar. The new race appears to resemble closely with race 77 as far as types of infection on the differential hosts are concerned. For comparative purposes the reactions of the newly isolated race of Puccinia triticina Eriks. and that of race 77 on the 8 differential varieties of wheat are set out in the following Table:—

Locality	Stock collection or isolation	Malakof	Carina	Brevit	Webster	Loros	Mediter	Hussar	Democrat
Pusa (Bihar)	Wheat Mediter- ranean	4	4	4	4	4	4	3-4	4
	Race 77	4	4	4	4	4	4	4	4

The new race is different from all other known Indian races of leaf rust as the 2 differential varieties, Mediterranean and Democrat are susceptible. The race was actually picked up from Mediterranean variety which had been sown at Pusa (Bihar).—Division of Mycology and Plant Pathology, I.A.R.I., New Delhi.

Plagionema Subramanian and Ramakrishnan, a synonym of Ciliochorella Sydow—B. L. Chona and R. L. Munjal. Ciliochorella Syd. is based on a monotypic collection of this fungus on Mangifera indica Linn. from Allahabad, India. The genus was described by Sydow, in Sydow and Mitter (1935), as one belonging to Sphaeropsidales, which lies close to Ciliochora and Diachorella Hoehnel but differs from these in its typical Phyllachoroid pycnidium and peculiar arrangement of the appendages of the spore.

Recently Subramanian and Ramakrishnan (1953) have described a new genus *Plagionema*, which agrees largely to the recorded description of *Ciliochorella* (Fig. 1). We had an opportunity to examine the Type material of both these genera and find them to be identical.

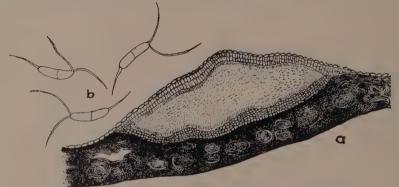


Fig. 1 - (a) T.S.through the leaf showing pycnidium of Ciliochorella mangiferae Sydow (From Type material) X 32 (b) Pycnospores X 750

Strangely enough, Subramanian and Ramakrishnan did not discuss the affinities of their newly named genus with other known related fungi, particularly when they collected this fungus on *Mangifera indica* as well, on which there is an earlier record of *Ciliochorella* from this country. The points of agreement and differences, as stated by the authors of the two genera and as made out by us after the study of Type materials of both, are discussed here.

Texture of fructifications has been vividly described in detail in the generic diagnosis of Ciliochorella by Sydow, which tallies exactly with that reported by Subramanian and Ramakrishnan for Plagionema. The development of the conidium, has been nicely followed up by Subramanian and Ramakrishnan and morphologically the conidia in the two genera are similar, though their interpretations are different. Possibly due to the dense granular nature of the protoplasmic contents of the spore, which imparts it a sub-hyaline colour, Sydow failed to report the central septum which is rather inconspicuous. Furthermore he described the basal appendage as stalk which is not correct, though outwardly it does appear to be so.

We have not observed any conidiophores bearing conidia. Rather conidia are borne on short elliptic cells of the basal stroma.

These cells put forth small papillae on which the development of conidia starts. We agree to the manner of development of the conidium as described by Subramanian and Ramakrishnan except that we have also seen basal appendage being formed on the short elliptic hyaline cell of the basal stroma. The basal appendage after slight development tapers down as a delicate hairy projection and is soon cut off, before the spore is ejected into the pycnidial cavity. We find it hard to agree with the authors of *Plagionema*, that the spores are 3 septate or 4 celled and feel that this may mislead other taxonomic workers. We regard the apical and basal cell-like outgrowths as protuberances of upper and lower cell of the spore, because these are very small in size as compared to the normal cell, are deviod of any protoplasmic contents and further grow out into delicate hair like appendages. We, thus, consider the spore as bicelled, with apical and basal appenages. We have also observed the abnormal conidia as described by Subramanian and Ramakrishnan in both the specimens of Ciliochorella mangiferae and Plagionema indica and rarely even 3celled spores.

We have made a number of collections of this fungus regularly at I. A. R. I. Delhi since 1948, on dead leaves of Syzygium jambos Alston and once on Mangifera indica Linn. and find that their measurement of spores, pycnidia etc., as well as those of collections of Plagionema indica agree with those of Ciliochorella mangiferae Syd. We, therefore, propose that the genus Plagionema together with its Type species P. indica be considered a synonym of Ciliochorella mangiferae Syd.

Ciliochorella bambusae described by Shanor from U.S. A. is a misleading record. That fungus has conidia different from Ciliochorella and also is characterised by the formation of distinct necrotic spots on living leaves, while C. mangiferae, the Type species, develops on dead leaves.

As the fungus has got bicelled, hyaline spores with dimidiate pycnidia and a shield above, we further propose that this be considered as one belonging to Hyalodidymiae of the family Leptostromaceae.

We are grateful to Dr. R. K. Saksena and Prof. T. S. Sadasivan for supplying the Type specimens. Our sincere thanks are due to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology for helpful criticism, encouragement and providing necessary facilities for this work.—Division of Mycology & Plant Pathology, Indian Agricultural Research Institute, New Delhi.

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J. H. Mitter

Chlorosis of Salvia coccinia Linn.—G. S. Verma & A. K. Bose. During the late winter of 1952, a few garden Salvia were observed with abnormal spotting on the leaves, very different from the normal healthy green leaves of unaffected plants in the beds. Investigations were carried out to determine the cause of the disease. Virus infection was suspected since neither any fungus nor bacteria could be isolated from the diseased leaves. The most consistent and visible symptoms invariably appeared in the foliage. The first visible manifestation of the disease appeared in the form of bright yellow spots of varying size on the leaf surface, preceded by clearing of the veins. The spots continued to enlarge, later coalesced to form continuous yellow patches and finally extended to give yellowish green to pale yellow colour to the entire leaf (fig. 1 A). This general chlorosis of the leaves is subsequently accompanied by such changes in the plants, as dwarfing, distortion, leaf curling and general stunting of the plants. (fig. 2).

The disease was not transmissible to any other host through mechanical inoculations. It was, however, readily transmissible to healthy plants by bud and cleft grafts, the latter being more successful. In the grafting experiments the disease was found to appear in the healthy plants, in about three weeks' time and in the form of yellow spots. There was, however, no noticeable distortion of leaves or stunting in grafted plants. The virus could not be transmitted to tomato and tobacco by grafting.

Gardner, Tompkins and Whipple (1935) reported that Salvia sp. was a host of tomato spotted wilt virus in California. Fulton (1941) isolated tobacco ring-spot virus from the roots of Salvia splendens, the leaves of which were previously inoculated with the virus. Holmes (1946) reported four species of Salvia as hosts of tobacco mosaic virus. Roland (1950) in Belgium observed a graft transmissible virus of Salvia splendens, which resembles the virus under study in Symptomatology. A search for the insect vector is in progress. This may reveal its correct identity.—Botany Department, Lucknow University, Lucknow.

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EXPLANATION OF THE PLATES

Fig. 1-A. Diseased Leaf. Fig. 1-B. Healthy Leaf. Fig. 2. Diseased Plant.

^{*}Original paper not consulted (Seen in Rev. App. Myc. 1951).







F--- 0

Two New Species of Synchtrium-S. C. Gupta and S. Sinha. During the rainy season of 1954 two new species of Synchytrium have been collected from fields in vicinity of Agra. The species concept as outlined by Mhatre & Mundkur (1945) and Gupta & Sinha (1951) has been followed. The specimens of the species reported here are deposited in the Herbarium of the Botany Department, Agra College, Agra and the Herb. Crypt. Ind. Orient. of Indian Agricultural Research Institute, New Delhi,

SYNCHYTRIUM CELOSIAE Gupta & Sinha, sp. nov-

Gallae in foliis, sparse, maxime singulares, globase, diametro 0.2=0.38 mm. Hypnospores singulares in cellis hostibus epidermatibus. globase, leves, fuse subnigrae, magnitudine 90=120 μ (aestimatio media 103.5μ) diametro cum episporio $9.0-11.6 \mu$ (aestimatio media 10.3μ) crasso.

In foliis Celosia argentea Linn. sp. Agra, 5-9-1954, leg. S. C. Gupta, typus.

Galls on leaves, sparsely distributed, mostly single, spherical, measuring 0.2-0.38 mm. (mean 0.3 mm.) in diameter, Resting sporangia solitary in epidermal cells, spherical, smooth, dark brown, measuring 90-120 μ (mean 103.5 μ) in diameter; epispore 9-11.6 μ (mean 10.3μ) thick.

On leaves of Celosia argentea Linn. Agra, 5-9-1954, leg. S. C. Gupta, type.

SYNCHYTRIUM CYAMOPSAE Gupta & Sinha, sp. nov.

Gallae in culmis atque foliis, maxime constantius venae, globase, diametro 0.23–0.48 mm. (aestimatio media 0.35 mm). Hypnospore globose, leves, fuse subnigre, maxime singulares, aliquando 2-3 in quisque galle, magnitudne $68-120~\mu$ (aestimato media 85.5μ) diametro cum episporio 1.7-6.6 \(\mu\) (aestimatio medida 4.6 \(\mu\)) crasso.

In culmis atque foliis Cyamopsis psoralioides D.C. Agra, 5-9-1954, leg. S. C. Gupta, typus.

Galls scattered on stem and leaves particularly along veins, measuring 0.23-0.48 mm. (mean 0.35 mm.) in diameter. Resting sporangia spherical, smooth, dark brown, mostly solitary sometimes 2-3 in each gall, measuring $68-120~\mu$ (mean $85.5~\mu$) in diameter; sporangial wall $1.7-6.6 \mu$ (mean 4.6μ) thick.

On stema nd leaves of Cyamopsis psoralioides D.C. Agra, 5-9-1954, leg. S. C. Gupta, type.—Botany Department, Agra College, Agra.

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Mutation in Puccinia graminis tritici (Pers.) Eriks. & Henn. Physiologic Race 15-C--D. P. Misra and V. C. Lele. During the normal work of maintenance of races at the Rust Research Laboratory, Simla, a dull-orange coloured pustule was observed in pure culture of race 15-C of Puccinia graminis tritici (Pers.) Eriks. & Henn. which has been found to differ from the type race in certain respects. The race was originally picked up by Gokhale (1950) and is being maintained at Simla since 14. 8. 1952. The mutant was observed in 15th generation at Simla Laboratory. Although cases of mutation in pathogenicity in rusts are rare, several cases of colour mutation have been reported from other countries. The mutant in P. g. tritici described here does not show any appreciable departure from the type race in pathogenicity but is distinct in morphological characters.

Uredospores of the mutant are dull-orange in colour and the epidermis of the uredosori does not rupture easily whereas that of race 15-C are dark brown in colour and the epidermis ruptures soon after the appearance of the pustule. The orange colour of the spores may be due to the absence of pigment in the spore wall as suggested by Newton, Johnson and Brown (1930). Besides the difference in colour, the spores differ in size as shown by the data set out in Table 1.

It is clear from the data that the uredospores of the mutant are smaller in size and that the differences are statistically significant; the value of 't' according to Fisher at 1% level for 100 observations being 2.57.

Germination tests with uredospores were conducted at temperature ranges of 39-45°, 50-62°, 70-80°, 77-83°, and 88-95°F. Temperature ranges upto 70-80°F, have been found to be congenial for germination of the type culture and the mutant but only 5% germination was observed at 77-83°F-range. No germination of spores of either of them was observed at 88-95°F. Significant differences in the percentage of germination of the type race and its mutant have only been observed at congenial temperatures and the percentage of germination as also rate of growth of the germ-tube of the mutant have been found to be consistently less than the type race.

Thirty-seven wheat and barley varieties including all the regular differentials of black rust and a few of brown and yellow rusts of wheat were selected for comparing the pathogenicity of the type race and the mutant. No difference in the pathogenicity of the type race and its mutant was observed. At higher range of temperature (40–87°F.) the mutant was, however, found to have a longer incubation period than the type race just by a day; and at lower range (39-76°F.) it was longer by three days, values at both ranges being significant. This was observed to be true in all the varieties under test.

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TABLE 1

Value of 't' (100 observations)	16.04	2.8447		
P. g. tritici uredospores of mutant. (Av. of 100)	28.35μ 15.75μ 22.52μ $\pm 2.8535\mu$ 0.2853μ 12.23%	18.90μ 12.60μ 15.68μ $\pm 0.9207\mu$ 0.0920μ 5.8%		
P. g. tritici uredospores of type race 15-C (Av. of 100)	34.65μ 18.90μ 28.76μ $\pm 2.6412\mu$ 0.2641μ 9.18%	18.90μ 13.86μ 16.01μ $\pm 0.7100\mu$ 0.0710μ 4.34%		
Standard statistical values.	Range Minimum Mean Standard Error Co-efficient of variability	Range (Maximum Mean Standard Deviation Standard Error Co-efficient of variability		
Dimensions of spores	Length of Uredospores.	Breadth of Uredospores.		

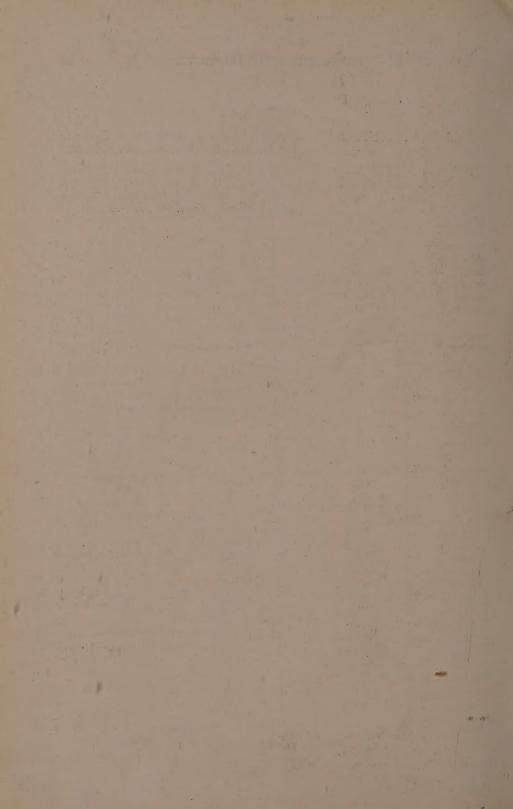
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